7-10-03

09718998

L4 ANSWER 1 OF 14 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2002:461308 CAPLUS
DOCUMENT NUMBER: 137:46047
TITLE: Method for making ***humanized*** antibodies
INVENTOR(S): Carter, Paul J.; Presta, Leonard G.
PATENT ASSIGNEE(S): Genentech, Inc., USA
SOURCE: U.S., 60 pp., Cont.-in-part of U.S. Ser. No. 715,272, abandoned.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 4
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6407213	B1	20020618	US 1993-146206	19931117
WO 9222653	A1	19921223	WO 1992-US5126	19920615
W: AU, CA, JP, US				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LU, MC, NL, SE				
EP 940468	A1	19990908	EP 1999-105252	19920615
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC				

PRIORITY APPLN. INFO.: US 1991-715272 B2 19910614
WO 1992-US5126 W 19920615
EP 1992-914220 A3 19920615

AB Variant Igs, particularly ***humanized*** antibody polypeptides are provided, along with methods for their prepn. and use. Consensus Ig sequences and structural models are utilized in this method.

REFERENCE COUNT: 265 THERE ARE 265 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 2 OF 14 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2000:401866 CAPLUS
DOCUMENT NUMBER: 133:39129
TITLE: Fluorescent proteins and their genes and muteins from non-bioluminescent species of class Anthozoa
INVENTOR(S): Lukyanov, Sergey Anatolievich; Fradkov, Arcady Fedorovich; Labas, Yulii Aleksandrovich; Matz, Mikhail Vladimirovich; Green, Gisele; Chen, Ying; Ding, Li
PATENT ASSIGNEE(S): Clontech Laboratories, Inc., USA
SOURCE: PCT Int. Appl., 86 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 15
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000034326	A1	20000615	WO 1999-US29473	19991210
W: JP, US				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				

PRIORITY APPLN. INFO.: US 1998-210330 A 19981211
US 1999-418529 A 19991014

AB The present invention is directed to novel fluorescent proteins from non-bioluminescent organisms from the Class Anthozoa. Hybridization probes were designed from consensus regions of known fluorescent proteins and used to isolate cDNA clones encoding fluorescent proteins from representative species. The cDNA sequence encoding the fluorescent protein (drFP583) from Discosoma sp. "red" is provided. The spectral properties of drFP583 are useful for labeling. ***Humanized*** DNA encoding drFP583 is constructed in which the codons are optimized for expression in mammalian cells, and site-specific oligonucleotide-directed mutagenesis or shuffling is applied to obtain variant and hybrid proteins with useful spectral properties.

REFERENCE COUNT: 5 THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 3 OF 14 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2000:796130 CAPLUS
TITLE: Discovery of a novel binding epitope for the .alpha.4 integrin based on sequence homology between VCAM-1 and the third heavy chain complementary-determining region of anti-.alpha.4 antibodies.
AUTHOR(S): Pleiss, Michael A.; Thorsett, Eugene D.; Yednock, Ted
CORPORATE SOURCE: Chemistry, Elan Pharmaceuticals, South San Francisco, CA, 94080, USA
SOURCE: Abstracts of Papers - American Chemical Society (2000), 220th, MEDI-132
CODEN: ACSRAL; ISSN: 0065-7727
PUBLISHER: American Chemical Society
DOCUMENT TYPE: Journal; Meeting Abstract
LANGUAGE: English
AB During the development of Antegren, a ***humanized*** anti-.alpha.4 antibody currently in Phase II clin. trials for multiple sclerosis, IBD and Crohnapos disease, it became necessary to sequence five murine anti-.alpha.4 antibodies as candidates for humanization. Comparison of the heavy chain CDR3 regions of these antibodies demonstrated a striking sequence homol. between four of the five antibodies: Y/FGN...Y. The fact that this motif is found in the CDR3 of four of the five antibodies suggests that it was probably important in integrin recognition since this occurrence was probably not a random coincidence. This was supported by the homol. seen with VCAM-1, the major adhesion partner for VLA4. Domain 1 of VCAM-1 is known to support adhesion with VLA4. A novel sequence (FGN&133;Y) is found in this domain, which has high identity with the sequence found in the CDR3 of .alpha.4. We have utilized this novel ***consensus*** ***sequence*** as a starting point for the rational design of small mol. inhibitors of VLA4. This presentation will deal with our efforts at demonstrating that this finding was valid by prepg. the 11-mer amino acid sequence from the CDR3 of AN100226m (the murine anti-.alpha.4 antibody that was ***humanized*** into Antegren) and showing that it was a weak but specific inhibitor of VLA4 specific adhesion. Utilization of classic peptide techniques (ala scans, eptitope mapping, cyclicizations, etc.) resulted in the prepn. of potent, cyclic hexapeptides that served as a starting point for partial mimetics and eventually small mol. inhibitors. The SAR developed during the course of this research will be presented.

L4 ANSWER 4 OF 14 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 2001:71739 BIOSIS
DOCUMENT NUMBER: PREV200100071739
TITLE: Discovery of a novel binding epitope for the alpha4 integrin based on sequence homology between vcam-1 and the third heavy chain complementary-determining region of anti-alpha4 antibodies.
AUTHOR(S): Pleiss, M. A. (1); Freedman, S. B.; Thorsett, E. D.; Yednock, T.
CORPORATE SOURCE: (1) Elan Pharmaceuticals, Inc., South San Francisco, CA USA
SOURCE: Society for Neuroscience Abstracts, (2000) Vol. 26, No. 1-2, pp. Abstract No.-126.18. print.
Meeting Info.: 30th Annual Meeting of the Society of Neuroscience New Orleans, LA, USA November 04-09, 2000 Society for Neuroscience
. ISSN: 0190-5295.
DOCUMENT TYPE: Conference
LANGUAGE: English
SUMMARY LANGUAGE: English
AB During the development of Antegren(R), a ***humanized*** anti-alpha4 antibody currently in Phase II clinical trials for multiple sclerosis, IBD and Crohn's disease, it became necessary to sequence five murine anti-alpha4 antibodies as candidates for humanization. Comparison of the heavy chain CDR3 regions of these antibodies demonstrated a striking

sequence homology between four of the five antibodies: Y/FGN...Y. The fact that this motif is found in the CDR3 of four of the five antibodies suggests that it was probably important in integrin recognition since this occurrence was probably not a random coincidence. This was supported by the homology seen with VCAM-1, the major adhesion partner for VLA4. Domain 1 of VCAM-1 is known to support adhesion with VLA4. A novel sequence (FGN...Y) is found in this domain, which has high identity with the sequence found in the CDR3 of alpha4. We have utilized this novel ***consensus*** ***sequence*** as a starting point for the rational design of small molecule inhibitors of VLA4. This presentation will deal with our efforts at demonstrating that this finding was valid by preparing the 11-mer amino acid sequence from the CDR3 of AN100226m (the murine anti-alpha4 antibody that was ***humanized*** into Antegren) and showing that it was a weak but specific inhibitor of VLA4 specific adhesion. Utilization of classic peptide techniques (ala scans, eptitope mapping, cyclicizations, etc.) resulted in the preparation of potent, cyclic hexapeptides that served as a starting point for partial mimetics and eventually small molecule inhibitors of this integrin.

L4 ANSWER 5 OF 14 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1998:635290 CAPLUS
DOCUMENT NUMBER: 129:314974
TITLE: Recombinant preparation of human IgG-type antibody containing grafted murine CDR to ganglioside GM2 for clinical use
INVENTOR(S): Nakamura, Kazuyasu; Hanai, Nobuo
PATENT ASSIGNEE(S): Kyowa Hakko Kogyo Co., Ltd., Japan
SOURCE: Jpn. Kokai Tokkyo Koho, 66 pp.
CODEN: JKXXAF
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 10257893	A2	19980929	JP 1997-66981	19970319
CA 2226400	AA	19980919	CA 1998-2226400	19980319
AU 9859420	A1	19981001	AU 1998-59420	19980319
AU 751948	B2	20020905		
EP 882794	A2	19981209	EP 1998-105047	19980319
EP 882794	A3	19990616		

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, MC, PT, IE, SI, LT, LV, FI, RO

PRIORITY APPLN. INFO.: JP 1997-66981 A 19970319
AB Disclosed is a method of recombinant prepn. in animal cells of a ***reshaped*** human IgG-type antibody contg. grafted murine CDR to ganglioside GM2, which antibody exhibits reduced side effects while maintains the same level of GM2-binding specificity and anti-tumor activities as does the ***humanized*** chimeric monoclonal antibody. The ***reshaped*** human antibody is prepd. by replacing the complementary detg. region (CDR) on VH and VL chains with the CDR from different antibodies. The ***reshaped*** human antibody also contains HMHCS (human most homologous ***consensus*** ***sequence***) from .gtoreq.1 of the frameworks (FR) of VH or VL of (non)human origin, which FR may also exhibit substitution mutation(s) at defined position(s). Claimed are (1) prepn. of expression plasmids based on tandem plasmid vector pKANTEX93; (2) prepn. of monoclonal antibodies KM8966, KM8967, KM8970, and KM8969, all contain the C.gamma.1 and C.kappa. chains from human and the CDR from mouse anti-human GM2 monoclonal antibody KM796; and (3) their use as diagnostic or anti-tumor agents against GM2-pos. cell.

L4 ANSWER 6 OF 14 MEDLINE
ACCESSION NUMBER: 1998265078 MEDLINE
DOCUMENT NUMBER: 98265078 PubMed ID: 9602368
TITLE: Somatic origin of T-cell epitopes within antibody variable regions: significance to monoclonal therapy and genesis of

systemic autoimmune disease.

AUTHOR: Wysocki L J; Zhang X; Smith D S; Snyder C M; Bonorino C
CORPORATE SOURCE: Department of Pediatrics, National Jewish Medical and Research Center, Denver, CO 80207, USA.. WysockiL@njc.org

CONTRACT NUMBER: R01AI39563 (NIAID)
SOURCE: IMMUNOLOGICAL REVIEWS, (1998 Apr) 162 233-46. Ref: 83
Journal code: 7702118. ISSN: 0105-2896.
PUB. COUNTRY: Denmark
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199808
ENTRY DATE: Entered STN: 19980820
Last Updated on STN: 19980820
Entered Medline: 19980813

AB During an immune response, specific antibody variable region genes are diversified by a somatic point mutation process that generates de novo "foreign" V-region sequences. This creates an interesting problem in immune regulation because B cells are highly proficient at self-presenting V-region peptides in the context of class II MHC. Though our studies indicate that the corresponding T-cell repertoire attains a state of tolerance to germline-encoded antibody V-region diversity, it is presently unknown whether the same is true of mutationally generated diversity. On the basis of immunoregulatory considerations, we hypothesize that contact exclusion or tolerance normally precludes T cells from helping B cells via self-presented mutant V-region peptides. The lack of recurrent somatic mutations that create known T-cell epitopes in antibody V regions lends some support to this idea. In contrast, our studies of spontaneously autoreactive B cells in systemic autoimmune disease strongly suggest that precursors of such cells are recruited by T-cell help directed to self-presented mutant idiopeptides. Failures in tolerance or contact exclusion mechanisms may be responsible for this apparently abnormal event. In addition to their importance in immune regulation, somatic mutations or other differences from germline-encoded V-region sequence may be largely responsible for undesirable patient responses to therapeutic monoclonal antibodies. These reactions might be averted or diminished by inducing tolerance in the T-cell repertoire with synthetic peptide correlates of non-germline-encoded V-region sequences in ***humanized*** antibodies.

L4 ANSWER 7 OF 14 MEDLINE

ACCESSION NUMBER: 1998021983 MEDLINE
DOCUMENT NUMBER: 98021983 PubMed ID: 9377574

TITLE: Humanization of an anti-vascular endothelial growth factor monoclonal antibody for the therapy of solid tumors and other disorders.

AUTHOR: Presta L G; Chen H; O'Connor S J; Chisholm V; Meng Y G; Krummen L; Winkler M; Ferrara N
CORPORATE SOURCE: Department of Immunology, Genentech, Inc., South San Francisco, California 94080, USA.

SOURCE: CANCER RESEARCH, (1997 Oct 15) 57 (20) 4593-9.
Journal code: 2984705R. ISSN: 0008-5472.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199711
ENTRY DATE: Entered STN: 19971224
Last Updated on STN: 19971224
Entered Medline: 19971110

AB Vascular endothelial growth factor (VEGF) is a major mediator of angiogenesis associated with tumors and other pathological conditions, including proliferative diabetic retinopathy and age-related macular degeneration. The murine anti-human VEGF monoclonal antibody (muMAb VEGF) A.4.6.1 has been shown to potently suppress angiogenesis and growth in a

variety of human tumor cells lines transplanted in nude mice and also to inhibit neovascularization in a primate model of ischemic retinal disease. In this report, we describe the humanization of muMAb VEGF A.4.6.1. by site-directed mutagenesis of a human framework. Not only the residues involved in the six complementarity-determining regions but also several framework residues were changed from human to murine. ***Humanized*** anti-VEGF F(ab) and IgG1 variants bind VEGF with affinity very similar to that of the original murine antibody. Furthermore, recombinant ***humanized*** MAb VEGF inhibits VEGF-induced proliferation of endothelial cells in vitro and tumor growth in vivo with potency and efficacy very similar to those of muMAb VEGF A.4.6.1. Therefore, recombinant ***humanized*** MAb VEGF is suitable to test the hypothesis that inhibition of VEGF-induced angiogenesis is a valid strategy for the treatment of solid tumors and other disorders in humans.

L4 ANSWER 8 OF 14 MEDLINE
ACCESSION NUMBER: 97248673 MEDLINE
DOCUMENT NUMBER: 97248673 PubMed ID: 9092614
TITLE: Compilation and analysis of intein sequences.
AUTHOR: Perler F B; Olsen G J; Adam E
CORPORATE SOURCE: New England Biolabs Inc., Beverly, MA 01915, USA.. perler@neb.com
SOURCE: NUCLEIC ACIDS RESEARCH, (1997 Mar 15) 25 (6) 1087-93. Journal code: 0411011. ISSN: 0305-1048.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199704
ENTRY DATE: Entered STN: 19970507
Last Updated on STN: 19970507
Entered Medline: 19970430

AB We have compiled a list of all the inteins (protein splicing elements) whose sequences have been published or were available from on-line sequence databases as of September 18, 1996. Analysis of the 36 available intein sequences refines the previously described intein motifs and reveals the presence of another intein motif, Block H. Furthermore, analysis of the new inteins ***reshapes*** our view of the conserved splice junction residues, since three inteins lack the intein penultimate His seen in prior examples. Comparison of intein sequences suggests that, in general, (i) inteins present in the same location within extein homologs from different organisms are very closely related to each other in paired sequence comparison or phylogenetic analysis and we suggest that they should be considered intein alleles; (ii) multiple inteins present in the same gene are no more similar to each other than to inteins present in different genes; (iii) phylogenetic analysis indicates that inteins are so divergent that trees with statistically significant branches cannot be generated except for intein alleles.

L4 ANSWER 9 OF 14 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 96075433 MEDLINE
DOCUMENT NUMBER: 96075433 PubMed ID: 7493379
TITLE: Designing human consensus antibodies with minimal positional templates.
AUTHOR: Couto J R; Christian R B; Peterson J A; Ceriani R L
CORPORATE SOURCE: Cancer Research Fund of Contra Costa, Walnut Creek, California 94596, USA.
SOURCE: CANCER RESEARCH, (1995 Dec 1) 55 (23 Suppl) 5973s-5977s. Journal code: 2984705R. ISSN: 0008-5472.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199601
ENTRY DATE: Entered STN: 19960217
Last Updated on STN: 19960217
Entered Medline: 19960111

AB A ***humanized*** antibody retains from the original murine antibody the variable region amino acid residues that are required for antigen binding. These generally include the grafted complementarity determining regions, as well as a few key framework residues. Although the remainder of the framework sequences are imported from a human antibody, they nevertheless differ at a few positions from the human consensus sequences. These atypical residues, which arose by somatic mutation during the affinity maturation of the chosen human antibody, could elicit an immune response in some of the patients receiving the ***humanized*** antibody. Thus, ideally one should, instead, choose human consensus frameworks for ***humanizing*** murine antibodies. Because there is a different ***consensus*** ***sequence*** for each of the subclasses of variable light and heavy chains, a method is needed to choose the most appropriate one. We are developing a minimal positional template for such a purpose. A minimal positional template indicates which positions in the variable region frameworks are absolutely required for maintaining the integrity of the binding domains. Therefore, to choose a human framework for humanization, one screens the available human consensus sequences for those that are most similar to the original murine sequence, specifically at the positions indicated by the template. In the subsequent humanization protocol, one then retains all of the murine residues found in the positions indicated in the template while ***humanizing*** the residues found at all other positions. A conservative positional template has been applied to the humanizations of the antibreast epithelial mucin antibodies BrE-3 and KC4-G3 without loss of binding affinity. Now we are using progressive cycles of computer modeling and laboratory testing to develop a minimal template. The first of such cycles produced template B, which has been used successfully in the humanization of the antibreast epithelial antigen BA46 antibody Mc3. This prompted us to design template C, which further approaches the desired minimal template. Future constructs will test the validity of this template as well as the validity of this novel humanization approach.

L4 ANSWER 10 OF 14 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1995:969304 CAPLUS
DOCUMENT NUMBER: 124:6714
TITLE: Designing human consensus antibodies with minimal positional templates
AUTHOR(S): Couto, Joseph R.; Christian, Rosemarie B.; Peterson, Jerry A.; Ceriani, Roberto L.
CORPORATE SOURCE: Cancer Research Fund of Contra Costa, Walnut Creek, CA, 94596, USA
SOURCE: Cancer Research (1995), 55(23, Suppl.), 5973S-7S
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A ***humanized*** antibody retains from the original murine antibody the variable region amino acid residues that are required for antigen binding. These generally include the grafted complementarity detg. regions, as well as a few key framework residues. Although the remainder of the framework sequences are imported from a human antibody, they nevertheless differ at a few positions from the human consensus sequences. These atypical residues, which arose by somatic mutation during the affinity maturation of the chosen human antibody, could elicit an immune response in some of the patients receiving the ***humanized*** antibody. Thus, ideally one should, instead, choose human consensus frameworks for ***humanizing*** murine antibodies. Because there is a different ***consensus*** ***sequence*** for each of the subclasses of variable light and heavy chains, a method is needed to choose the most appropriate one. The authors are developing a minimal positional template for such a purpose. A minimal positional template indicates which positions in the variable region frameworks are absolutely required for maintaining the integrity of the binding domains. Therefore, to choose a human framework for humanization, one screens the available human consensus sequences for those that are more similar to the original murine sequence, specifically at the positions indicated by the template.

In the subsequent humanization protocol, one then retains all of the murine residues found in the positions indicated in the template while ***humanizing*** the residues found at all other positions. A conservative positional template has been applied to the humanizations of the antibreast epithelial mucin antibodies BrE-3 and KC4-G3 without loss of binding affinity. Now the authors are using progressive cycles of computer modeling and lab. testing to develop a minimal template. The first of such cycles produced template B, which has been used successfully in the humanization of the antibreast epithelial antigen BA46 antibody Mc3. This prompted the authors to design template C, which further approaches the desired minimal template. Future constructs will test the validity of this template as well as the validity of this novel humanization approach.

L4 ANSWER 11 OF 14 MEDLINE
ACCESSION NUMBER: 95228057 MEDLINE
DOCUMENT NUMBER: 95228057 PubMed ID: 7712480
TITLE: Anti-BA46 monoclonal antibody Mc3: humanization using a novel positional consensus and in vivo and in vitro characterization.
AUTHOR: Couto J R; Blank E W; Peterson J A; Ceriani R L
CORPORATE SOURCE: Cancer Research Fund of Contra Costa, Walnut Creek, California 94596, USA.
SOURCE: CANCER RESEARCH, (1995 Apr 15) 55 (8) 1717-22.
Journal code: 2984705R. ISSN: 0008-5472.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199505
ENTRY DATE: Entered STN: 19950524
Last Updated on STN: 19950524
Entered Medline: 19950515

AB Mc3 is a murine mAb that is highly effective in treating breast tumors in experimental radioimmunotherapy. Mc3 binds to BA46, a 46-kDa glycoprotein of the human milk fat globule membrane that is also expressed in breast carcinoma cells. We cloned and sequenced cDNAs encoding the variable regions of Mc3 and constructed an IgG1, kappa human/mouse chimeric antibody. We then ***humanized*** the variable regions of Mc3 using a positional consensus method and retaining residues that might either contact the complementarity-determining regions or the opposite chain. This positional consensus is novel in that it does not include residues with buried side chains. ***Humanized*** Mc3 retained full binding affinity, and fully competes with murine Mc3 for antigen binding. ***Humanized*** and murine 131I-labeled Mc3 behaved identically in athymic nu/nu mice biodistribution studies. The tumor uptake levels for both antibodies increased over a period of 4 days within a range of 13-20% of the injected dose per g with extremely favorable tumor:normal ratios. Also, a single therapeutic dose of 131I-labeled ***humanized*** Mc3 in the same animal model reduced the average tumor size and produced one of five cures while in the uninjected control tumor growth continued unabated. We believe that these results justify the implementation of Phase I human clinical trials for imaging and radioimmunotherapy of breast cancer.

L4 ANSWER 12 OF 14 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1993:624264 CAPLUS
DOCUMENT NUMBER: 119:224264
TITLE: Methods and materials for preparation of modified antibody variable domains and therapeutic uses thereof
INVENTOR(S): Studnicka, Gary M.; Little, Roger G., II; Fishwild, Dianne M.; Kohn, Fred R.
PATENT ASSIGNEE(S): Xoma Corp., USA
SOURCE: PCT Int. Appl., 159 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English

FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9311794	A1	19930624	WO 1992-US10906	19921214
W: CA, JP, US				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
CA 2103887	AA	19930614	CA 1992-2103887	19921214
EP 571613	A1	19931201	EP 1993-901238	19921214
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE				
JP 06506362	T2	19940721	JP 1993-511171	19921214
EP 1291360	A1	20030312	EP 2002-21775	19921214
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE				
US 5869619	A	19990209	US 1993-82842	19930623
US 5766886	A	19980616	US 1993-107669	19930813
US 5821123	A	19981013	US 1995-477531	19950607

PRIORITY APPLN. INFO.: US 1991-808464 A2 19911213
EP 1993-901238 A3 19921214
WO 1992-US10906 W 19921214
US 1993-107669 A1 19930813

AB Methods are described for identifying the amino acid residues of an antibody (Ab) variable domain which may be modified without diminishing the native affinity of the domain for antigen, while reducing its immunogenicity with respect to a heterologous species. Also described are methods for prepg. the so-modified Ab variable domains, which are useful for administration to heterologous species, and the Ab variable regions so prepd. The methodol. of the invention includes (1) detg. the amino acid sequences for light and heavy chain variable regions of the Ab to be modified; (2) aligning by homol. the light chain sequence with a plurality of human light chain sequences, and the heavy chain sequence with a plurality of human heavy chain sequences; (3) identifying amino acids in the light and heavy chain sequences which are least likely to diminish the affinity of the variable region for antigen while, at the same time, reducing its immunogenicity, by selecting each amino acid which is not in an interface region of the Ab variable domain and which is not in a complementarity-detg. region or in an antigen-binding region of the Ab variable domain, but which amino acid is in a position exposed to a solvent contg. the Ab; and (4) changing each amino acid identified in (3) which aligns with a highly or moderately conserved amino acid in the plurality of human light or heavy chain sequences if the identified amino acid is different from the amino acid in the plurality of human light and heavy chain sequences. The method of the invention was applied to modification of the variable region of murine monoclonal antibody (MAb) H65 (reactive with human CD5 antigen); comparative H65 and human sequences are included, as are sequences of the modified variable regions. Genes encoding ***humanized*** H65 light and heavy chain variable regions were constructed, and the ***humanized*** H65 Ab was expressed. The low-risk changes made in the course of modification of ***humanized*** H65 did not diminish the binding affinity of this Ab for the CD5 antigen. Also described are depletion of human T-cells from SCID mice by treatment with H65 MAb, the effects of a anti-Lyt-1 MAb (Lyt-1 is the murine equiv. of CD5) on lymphocytes and on collagen-induced arthritis in mice, etc.

L4 ANSWER 13 OF 14 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 93347389 EMBASE
DOCUMENT NUMBER: 1993347389
TITLE: Humanization of a mouse anti-human IgE antibody: A potential therapeutic for IgE-mediated allergies.
AUTHOR: Kolbinger F.; Saldanha J.; Hardman N.; Bendig M.M.
CORPORATE SOURCE: Med Resrch Council Collaborative Ctr, 1-3 Burtonhole Lane,Mill Hill, London NW7 1AD, United Kingdom
SOURCE: Protein Engineering, (1993) 6/8 (971-980).
ISSN: 0269-2139 CODEN: PRENE
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 026 Immunology, Serology and Transplantation

029 Clinical Biochemistry
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Mouse mAb TES-C21(C21) recognizes an epitope on human IgE and, therefore, has potential as a therapeutic agent in patients with IgE-mediated allergies such as hay fever, food and drug allergies and extrinsic asthma. The clinical usefulness of mouse antibodies is limited, however, due to their immunogenicity in humans. Mouse C21 antibody was ***humanized*** by complementarity determining region (CDR) grafting with the aim of developing an effective and safe therapeutic for the treatment of IgE-mediated allergies. The CDR-grafted, or ***reshaped*** human, C21 variable regions were carefully designed using a specially constructed molecular model of the mouse C21 variable regions. A key step in the design of ***reshaped*** human variable regions is the selection of the human framework regions (FRs) to serve as the backbones of the ***reshaped*** human variable regions. Two approaches to the selection of human FRs were tested: (i) selection from human consensus sequences and (ii) selection from individual human antibodies. The ***reshaped*** human and mouse C21 antibodies were tested and compared using a biosensor to measure the kinetics of binding to human IgE. Surprisingly, a few of the ***reshaped*** human C21 antibodies exhibited patterns of binding and affinities that were essentially identical to those of mouse C21 antibody.

L4 ANSWER 14 OF 14 MEDLINE
ACCESSION NUMBER: 92047485 MEDLINE
DOCUMENT NUMBER: 92047485 PubMed ID: 1943136
TITLE: Antigenicity of mouse monoclonal antibodies. A study on the variable region of the heavy chain.
AUTHOR: Olsson P G; Hammarstrom L; Smith C I
CORPORATE SOURCE: Department of Clinical Immunology, Karolinska Institute, Huddinge University Hospital, Sweden.
SOURCE: JOURNAL OF THEORETICAL BIOLOGY, (1991 Jul 7) 151 (1) 111-22.
Journal code: 0376342. ISSN: 0022-5193.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199111
ENTRY DATE: Entered STN: 19920124
Last Updated on STN: 19920124
Entered Medline: 19911125
AB Mouse monoclonal antibodies (Mabs) against human tumour antigens are currently used in therapy, but up to 50% of the patients receiving treatment form anti-Mab antibodies thus reducing the efficiency of the treatment. One attempt to minimize the immunogenicity of the mouse Mabs is to " ***humanize*** " them by replacing the constant part of the molecule with the human equivalent by genetic engineering. However, this does not reduce the immunogenicity of the variable part of the antibody. Some variable regions may be expected to be less antigenic than others. We therefore compared consensus sequences for the 11 mouse VH families with the human VH sequences published so far. Theoretical antigenicity predictions (hydrophilicity, flexibility, surface accessibility and relative antigenicity) were made and two families; VH I(J558) and VH XI (CP5 B5-3) were predicted to be immunogenic by all four methods. One family, VH X (MRL-DNA4), was not predicted to be immunogenic by any of the four methods. The residues predicted to form antigenic epitopes in the two families VH II (Q52) and VH III (36-60) are predicted not to be exposed on the surface of the antibody molecule and may therefore not be immunogenic.

L9 ANSWER 1 OF 7 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.DUPLICATE 1
ACCESSION NUMBER: 2001:348979 BIOSIS

DOCUMENT NUMBER: PREV200100348979
TITLE: ***Humanized*** immunoglobulins and methods of making the same.
AUTHOR(S): Queen, Cary L.; Selick, Harold E.
ASSIGNEE: Protein Design Labs, Inc.
PATENT INFORMATION: US 6180370 January 30, 2001
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (Jan. 30, 2001) Vol. 1242, No. 5, pp. No Pagination. e-file.
ISSN: 0098-1133.
DOCUMENT TYPE: Patent
LANGUAGE: English
AB Novel methods for producing, and compositions of, ***humanized*** immunoglobulins having one or more complementarity determining regions (CDR's) and possible additional amino acids from a donor immunoglobulin and a ***framework*** region from an accepting human immunoglobulin are provided. Each ***humanized*** immunoglobulin chain will usually comprise, in addition to the CDR's, amino acids from the donor immunoglobulin ***framework*** that are, e.g., capable of interacting with the CDR's to effect binding affinity, such as one or more amino acids which are immediately ***adjacent*** to a CDR in the donor immunoglobulin or those within about about 3 ANG as predicted by molecular modeling. The heavy and light chains may each be designed by using any one or all of various position criteria. When combined into an intact antibody, the ***humanized*** immunoglobulins of the present invention will be substantially non-immunogenic in humans and retain substantially the same affinity as the donor immunoglobulin to the antigen, such as a protein or other compound containing an epitope.

L9 ANSWER 2 OF 7 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 2001357851 MEDLINE
DOCUMENT NUMBER: 21311885 PubMed ID: 11418663
TITLE: Structural effects of ***framework*** mutations on a ***humanized*** anti-lysozyme antibody.
AUTHOR: Holmes M A; Buss T N; Foote J
CORPORATE SOURCE: Program in Molecular Medicine, Fred Hutchinson Cancer Research Center, Seattle, WA 98109, USA.
SOURCE: JOURNAL OF IMMUNOLOGY, (2001 Jul 1) 167 (1) 296-301.
Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
OTHER SOURCE: PDB-H27S; PDB-H71A; PDB-H71R; PDB-H71V
ENTRY MONTH: 200109
ENTRY DATE: Entered STN: 20010924
Last Updated on STN: 20010924
Entered Medline: 20010920
AB A ***humanized*** version of the mouse anti-lysozyme Ab D1.3 was previously constructed as an Fv fragment and its structure was crystallographically determined in the free form and in complex with lysozyme. Here we report five new crystal structures of single-amino acid substitution mutants of the ***humanized*** Fv fragment, four of which were determined as Fv-lysozyme complexes. The crystals were isomorphous with the parent forms, and were refined to free R values of 28-31% at resolutions of 2.7-2.9 A. Residue 27 in other Abs has been implicated in stabilizing the conformation of the first complementarity-determining region (CDR) of the H chain, residues 31-35. We find that a Phe-to-Ser mutation at 27 alters the conformation of immediately ***adjacent*** residues, but this change is only weakly transmitted to Ag binding residues in the nearby CDR. Residue 71 of the H chain has been proposed to control the relative disposition of H chain CDRs 1 and 2, based on the bulk of its side chain. However, in structures we determined with Val, Ala, or Arg substituted in place of Lys at position 71, no significant change in the conformation of CDRs 1 and 2 was observed.

L9 ANSWER 3 OF 7 MEDLINE DUPLICATE 3

ACCESSION NUMBER: 1999326368 MEDLINE
DOCUMENT NUMBER: 99326368 PubMed ID: 10398393
TITLE: Comparison of the three-dimensional structures of a ***humanized*** and a chimeric Fab of an anti-gamma-interferon antibody.
AUTHOR: Fan Z C; Shan L; Goldsteen B Z; Guddat L W; Thakur A; Landolfi N F; Co M S; Vasquez M; Queen C; Ramsland P A; Edmundson A B
CORPORATE SOURCE: Oklahoma Medical Research Foundation, 825 NE 13th Street, Oklahoma City, OK 73104, USA.
CONTRACT NUMBER: CA 72803 (NCI)
SOURCE: JOURNAL OF MOLECULAR RECOGNITION, (1999 Jan-Feb) 12 (1) 19-32. Ref: 78
Journal code: 9004580. ISSN: 0952-3499.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200003
ENTRY DATE: Entered STN: 20000320
Last Updated on STN: 20000320
Entered Medline: 20000309

AB The objective of this work is to compare the three-dimensional structures of " ***humanized*** " and mouse-human chimeric forms of a murine monoclonal antibody elicited against human gamma-interferon. It is also to provide structural explanations for the small differences in the affinities and biological interactions of the two molecules for this antigen. Antigen-binding fragments (Fabs) were produced by papain hydrolysis of the antibodies and crystallized with polyethylene glycol (PEG) 8,000 by nearly identical microseeding procedures. Their structures were solved by X-ray analyses at 2.9 A resolution, using molecular replacement methods and crystallographic refinement. Comparison of these structures revealed marked similarities in the light (L) chains and near identities of the constant (C) domains of the heavy (H) chains. However, the variable (V) domains of the heavy chains exhibited substantial differences in the conformations of all three complementarity-determining regions (CDRs), and in their first ***framework*** segments (FR1). In FR1 of the ***humanized*** VH, the substitution of serine for proline in position 7 allowed the N-terminal segment (designated strand 4-1) to be closely juxtaposed to an ***adjacent*** strand (4-2) and form hydrogen bonds ***typical*** of an antiparallel beta-pleated sheet. The tightening of the ***humanized*** structure was relayed in such a way as to decrease the space available for the last portion of HFR1 and the first part of HCDR1. This compression led to the formation of an alpha-helix involving residues 25-32. With fewer steric constraints, the corresponding segment in the chimeric Fab lengthened by at least 1 A to a random coil which terminated in a single turn of 310 helix. In the ***humanized*** Fab, HCDR1, which is sandwiched between HCDR2 and HCDR3, significantly influenced the structures of both regions. HCDR2 was forced into a bent and twisted orientation different from that in the chimeric Fab, both at the crown of the loop (around proline H52a) and at its base. As in HCDR1, the last few residues of HCDR2 in the ***humanized*** Fab were compressed into a space-saving alpha-helix, contrasting with a more extended 310 helix in the chimeric form. HCDR3 in the ***humanized*** Fab was also adjusted in shape and topography. The observed similarities in the functional binding activities of the two molecules can be rationalized by limited induced fit adjustments in their structures on antigen binding. While not perfect replicas, the two structures are testimonials to the progress in making high affinity monoclonal antibodies safe for human use.

L9 ANSWER 4 OF 7 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 1999030463 MEDLINE
DOCUMENT NUMBER: 99030463 PubMed ID: 9811544

TITLE: Crystal structures of a rat anti-CD52 (CAMPATH-1) therapeutic antibody Fab fragment and its ***humanized*** counterpart.
AUTHOR: Cheetham G M; Hale G; Waldmann H; Bloomer A C
CORPORATE SOURCE: MRC Laboratory of Molecular Biology, Hills Road, Cambridge, CB2 2QH, UK.
SOURCE: JOURNAL OF MOLECULAR BIOLOGY, (1998 Nov 20) 284 (1) 85-99. Journal code: 2985088R. ISSN: 0022-2836.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: PDB-1RBEY; PDB-R1BEYST
ENTRY MONTH: 199902
ENTRY DATE: Entered STN: 19990311
Last Updated on STN: 20030313
Entered Medline: 19990225

AB The CAMPATH-1 family of antibodies are able systematically to lyse human lymphocytes with human complement by targeting the small cell-surface glycoprotein CD52, commonly called the CAMPATH-1 antigen. These antibodies have been used clinically for several years, providing therapy for patients with a variety of immunologically mediated diseases. We report here the first X-ray crystallographic analyses of a Fab fragment from a rat antibody, the original therapeutic monoclonal CAMPATH-1G and its ***humanized*** counterpart CAMPATH-1H, into which the six complementarity-determining regions of the rat antibody have been introduced. These structures have been refined at 2.6 A and 3.25 A resolution, respectively. The VL domains of ***adjacent*** molecules of CAMPATH-1H form a symmetric dimer within the crystals with an inter-molecular extended beta-sheet as seen in light chain dimers of the kappa class. Crystals of CAMPATH-1G have translational pseudo-symmetry. Within the antibody-combining sites, which are dominated by the protrusion of LysH52b and LysH53 from hypervariable loop H2, the charge distribution and overall integrity are highly conserved, but large changes in the position of loop H1 are observed and an altered conformation of loop H2. The major determinants of this are ***framework*** residues H71 and H24, whose identity differs in these two antibodies. These structures provide a detailed structural insight into the transplantation of an intact antibody-combining site between a rodent and a human ***framework*** , and provide an increased understanding of the specificity and antigen affinity of this pair of CAMPATH-1 antibodies for CD52. This study forms the structural basis for future modification and design of more effective antibodies to this important antigen.

L9 ANSWER 5 OF 7 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1996:447099 CAPLUS
DOCUMENT NUMBER: 125:140550
TITLE: ***Humanized*** immunoglobulins
INVENTOR(S): Queen, Cary L.; Selick, Harold E.
PATENT ASSIGNEE(S): Protein Design Labs, Inc., USA
SOURCE: U.S., 139 pp., Cont.-in-part of U.S. Ser. No. 590,274, abandoned.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 4
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 5530101	A	19960625	US 1990-634278	19901219
ZA 8909956	A	19901031	ZA 1989-9956	19891228
EP 682040	A1	19951115	EP 1995-105609	19891228
EP 682040	B1	19990825		
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE				
JP 11004694	A2	19990112	JP 1998-4334	19891228

EP 939127 A2 19990901 EP 1998-204240 19891228
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE
CA 2328851 C 20020813 CA 1989-2328851 19891228
DD 296964 A5 19911219 DD 1990-337159 19900117
CA 2098404 AA 19920620 CA 1991-2098404 19911219
CA 2098404 C 20020820
WO 9211018 A1 19920709 WO 1991-US9711 19911219
W: AT, AU, BB, BG, BR, CA, CH, CS, DE, DK, ES, FI, GB, HU, JP, KP, KR, LK, LU, MG, MN, MW, NL, NO, PL, RO, SD, SE, SU
RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GN, GR, IT, LU, MC, ML, MR, NL, SE, SN, TD, TG
AU 9191726 A1 19920722 AU 1991-91726 19911219
AU 671949 B2 19960919
EP 566647 A1 19931027 EP 1992-903551 19911219
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, MC, NL, SE
JP 06503963 T2 19940512 JP 1991-503758 19911219
JP 3276369 B2 20020422 JP 1992-503758 19911219
US 5585089 A 19961217 US 1995-477728 19950607
US 5693761 A 19971202 US 1995-474040 19950607
US 5693762 A 19971202 US 1995-487200 19950607
US 6180370 B1 20010130 US 1995-484537 19950607
SG 78258 A1 20010220 SG 1996-7855 19960413
AU 9675481 A1 19970220 AU 1996-75481 19961219
PRIORITY APPLN. INFO.: US 1988-290975 B2 19881228
US 1989-310252 B2 19890213
US 1990-590274 B2 19900928
CA 1989-2006865 A3 19891228
EP 1990-903576 A3 19891228
EP 1995-105609 A3 19891228
JP 1990-503677 A3 19891228
US 1990-634278 A 19901219
WO 1991-US9711 A 19911219

AB Novel methods for producing, and compns. of, ***humanized*** Igs having one or more complementarity detg. regions (CDR's) and possible addnl. amino acids from a donor Ig and a ***framework*** region from an accepting human Ig are provided. Each ***humanized*** Ig chain will usually comprise, in addn. to the CDR's, amino acids from the donor Ig ***framework*** that are, e.g., capable of interacting with the CDR's to effect binding affinity, such as one or more amino acids which are immediately ***adjacent*** to a CDR in the donor Ig or those within about 3 .ANG. as predicted by mol. modeling. The heavy and light chains may each be designed by using any one or all of various position criteria. When combined into an intact antibody, the ***humanized*** Igs of the present invention will be substantially non-immunogenic in humans and retain substantially the same affinity as the donor Ig to the antigen, such as a protein or other compd. contg. an epitope. In example, prepd. were ***humanized*** antibodies specific for Tac (activated T cell), gB and gD glycoproteins of herpes simplex virus, CD33 antigen, p75 of interleukin 2 receptor, gH glycoprotein of cytomegalovirus.

L9 ANSWER 6 OF 7 MEDLINE DUPLICATE 5
ACCESSION NUMBER: 95232114 MEDLINE
DOCUMENT NUMBER: 95232114 PubMed ID: 7716162
TITLE: Efficient generation of a ***reshaped*** human mAb specific for the alpha toxin of Clostridium perfringens.
AUTHOR: Tempest P R; White P; Williamson E D; Titball R W; Kelly D C; Kemp G J; Gray P M; Forster S J; Carr F J; Harris W J
CORPORATE SOURCE: Scotgen Biopharmaceuticals Inc., Aberdeen, UK.
SOURCE: PROTEIN ENGINEERING, (1994 Dec) 7 (12) 1501-7.
Journal code: 8801484. ISSN: 0269-2139.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-L25352; GENBANK-L25353
ENTRY MONTH: 199505
ENTRY DATE: Entered STN: 19950524

Last Updated on STN: 19990129
Entered Medline: 19950516

AB We have used the technique of antibody ***reshaping*** to produce a ***humanized*** antibody specific for the alpha toxin of Clostridium perfringens. The starting antibody was from a mouse hybridoma from which variable (V) region nucleotide sequences were determined. The complementarity-determining regions (CDRs) from these V regions were then inserted into human heavy and light chain V region genes with human constant region gene fragments subsequently added. The insertion of CDRs alone into human frameworks did not produce a functional ***reshaped*** antibody and modifications to the V region ***framework*** were required. With minor ***framework*** modifications, the affinity of the original murine mAb was restored and even exceeded. Where affinity was increased, an altered binding profile to overlapping peptides was observed. Computer modelling of the ***reshaped*** heavy chain V regions suggested that amino acids ***adjacent*** to CDRs can either contribute to, or distort, CDR loop conformation and must be adjusted to achieve high binding affinity.

L9 ANSWER 7 OF 7 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1991:183735 CAPLUS
DOCUMENT NUMBER: 114:183735
TITLE: Chimeric immunoglobulins specific for p55 Tac protein of the interleukin-2 (IL-2) receptor
INVENTOR(S): Queen, Cary L.; Selick, Harold Edwin
PATENT ASSIGNEE(S): Protein Design Labs, Inc., USA
SOURCE: PCT Int. Appl., 48 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 4
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9007861	A1	19900726	WO 1989-US5857	19891228
W: AT, AU, BB, BG, BR, CH, DE, DK, FI, GB, HU, JP, KP, KR, LK, LU, MC, MG, MW, NL, NO, RO, SD, SE, SU				
RW: AT, BE, BF, BJ, CF, CG, CH, CM, DE, ES, FR, GA, GB, IT, LU, ML, MR, NL, SE, SN, TD, TG				
CA 2006865	AA	19900628	CA 1989-2006865	19891228
CA 2006865	C	20020820		
CN 1043875	A	19900718	CN 1989-109618	19891228
CN 1057013	B	20001004		
AU 9051532	A1	19900813	AU 1990-51532	19891228
AU 647383	B2	19940324		
ZA 8909956	A	19901031	ZA 1989-9956	19891228
EP 451216	A1	19911016	EP 1990-903576	19891228
EP 451216	B1	19960124		
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE				
JP 04502408	T2	19920507	JP 1990-503677	19891228
EP 682040	A1	19951115	EP 1995-105609	19891228
EP 682040	B1	19990825		
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE				
AT 133452	E	19960215	AT 1990-903576	19891228
ES 2081974	T3	19960316	ES 1990-903576	19891228
JP 11004694	A2	19990112	JP 1998-4334	19891228
RU 2126046	C1	19990210	RU 1989-4895847	19891228
EP 939127	A2	19990901	EP 1998-204240	19891228
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE				
AT 183753	E	19990915	AT 1995-105609	19891228
ES 2136760	T3	19991201	ES 1995-105609	19891228
CA 2328851	C	20020813	CA 1989-2328851	19891228
DD 296964	A5	19911219	DD 1990-337159	19900117
DK 9101191	A	19910619	DK 1991-1191	19910619
NO 9102385	A	19910619	NO 1991-2385	19910619
SG 78258	A1	20010220	SG 1996-7855	19960413

DK 9800941 A 19980716 DK 1998-941 19980716
DK 174317 B1 20021202
HK 1014718 A1 20000714 HK 1998-115967 19981228
PRIORITY APPLN. INFO.: US 1988-290975 A 19881228
US 1989-310252 A 19890213
CA 1989-2006865 A3 19891228
EP 1990-903576 A3 19891228
EP 1995-105609 A3 19891228
JP 1990-503677 A3 19891228
WO 1989-US5857 A 19891228

AB Methods for designing ***humanized*** Igs having .gtoreq.1 complementary detg. regions (CDRs) from a donor Ig and a ***framework*** region from a human Ig comprise 1st comparing the ***framework*** or variable region amino acid sequence of the donor Ig to corresponding sequences in a collection of human Ig chains and selecting as the human Ig .gtoreq. 1 homologous sequences from the collection. Each ***humanized*** Ig chain may comprise approx. .gtoreq.3 amino acids from the donor Ig in addn. to the CDRs, usually .gtoreq.1 of which is immediately ***adjacent*** to a CDR in the donor Ig. The heavy and light chains may each be designed by using any 1 or all 3 addnl. position criteria. When combined in an intact antibody, the ***humanized*** Igs of the invention will be substantially nonimmunogenic in humans and retain substantially the same affinity as the donor Ig to the antigen. The above method is applied to design and prodn. of the title chimeric antibodies. Thus, comparison of human antibody Eu and anti-Tac antibody sequences allowed design of genes for human-like light and heavy chains. Appropriate oligonucleotide segments were synthesized and annealed to form these genes, which were then used to construct plasmids pHuGTAC1 (for ***humanized*** heavy chain prodn.) and pHuLTAC (for ***humanized*** light chain prodn.). These 2 plasmids were transfected into mouse sp2/0 cells for prodn. of the title antibody. The secreted antibody bound to HUT-102 cells (which express the IL-2 receptor), but not to Jurkat T-cells (which do not express the IL-2 receptors). The ***humanized*** and original anti-Tac antibodies have approx. the same affinity (within 3-4-fold). In 51Cr-release antibody-dependent cell-mediated cytotoxicity assays using effector cell/target cell ratios of 30:1 or 100:1, anti-Tac antibody lysed <5% of target cells, while the ***humanized*** antibody lysed >20% of target cells.

L12 ANSWER 1 OF 34 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2003:472707 CAPLUS
TITLE: Screening and selection of immunoglobulin heavy chain ***CDR*** domains for display in microscaffolds
INVENTOR(S): Lasters, Ignace; Pletinckx, Jurgen; Boutonnet, Nathalie; Lauwereys, Marc; Beirnaert, Els
PATENT ASSIGNEE(S): Algonomics N.V., Belg.; Ablynx N.V.
SOURCE: PCT Int. Appl., 90 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003050531	A2	20030619	WO 2002-BE189	20021211

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL,

PT, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG
PRIORITY APPLN. INFO.: EP 2001-870274 A 20011211
AB The authors disclose polypeptide micro-scaffolds displaying Ig CDR2 or CDR3 polypeptide sequences. The micro-scaffolds comprise a CDR2 or CDR3 polypeptide sequence interconnecting fragments of the ***adjacent*** ***framework*** polypeptide sequences, which are arranged to form two anti-parallel .beta.-strands. The present invention is further related to a method to search, select or screen for Ig CDR2 or CDR3 polypeptide sequences that bind to a given antigen or mixt. of antigens, comprising the steps of: Creating a ***CDR*** library with the method of claim 13 from the genetic information of an individual or group of individuals; Select a ***CDR*** , which binds to said antigen or mixt. of antigens.

L12 ANSWER 2 OF 34 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2002:449724 CAPLUS
DOCUMENT NUMBER: 137:32068
TITLE: Rationally designed antibodies substituting ***CDR*** with hematopoietin peptide mimetic for diagnosis and therapy
INVENTOR(S): Bowdish, Katherine S.; Barbas-Frederickson, Shana; Renshaw, Mark
PATENT ASSIGNEE(S): Alexion Pharmaceuticals, Inc., USA
SOURCE: PCT Int. Appl., 113 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002046238	A2	20020613	WO 2001-US47656	20011205
AU 2002034001	A5	20020618	AU 2002-34001	20011205
US 2003049683	A1	20030313	US 2001-6593	20011205

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 2000-251448P P 20001205
US 2001-288889P P 20010504
US 2001-294068P P 20010529
WO 2001-US47656 W 20011205
AB Antibodies or fragments thereof having ***CDR*** regions replaced or fused with biol. active peptides are described. The antibody is human anti-tetanus toxoid antibody, and the biol. active peptide is selected from thrombopoietin peptide mimetic or erythropoietin peptide mimetic. The antibody-peptide fusion product also comprises flanking sequence(s) that may optionally be attached at one or both the carboxy-terminal and amino-terminal ends of the peptide in covalent assocn. with ***adjacent*** ***framework*** regions. Compns. contg. such antibody or fragment fusion products are useful in therapeutic and diagnostic modalities, e.g. for stimulating proliferation, differentiation, or growth of megakaryocytes, hematopoietic stem cells, or progenitors, and for increasing prodn. of platelets or red blood cells.

L12 ANSWER 3 OF 34 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 2002482298 MEDLINE
DOCUMENT NUMBER: 22229534 PubMed ID: 12244172
TITLE: Evidence for involvement of a hydrophobic patch in ***framework*** region 1 of human V4-34-encoded Igs in recognition of the red blood cell I antigen.

AUTHOR: Potter Kathleen N; Hobby Paul; Klijn Susanne; Stevenson Freda K; Sutton Brian J
CORPORATE SOURCE: Molecular Immunology Group, Tenovus Laboratory, Southampton University Hospitals Trust, Southampton, United Kingdom.. kp1@soton.ac.uk
SOURCE: JOURNAL OF IMMUNOLOGY, (2002 Oct 1) 169 (7) 3777-82.
Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200211
ENTRY DATE: Entered STN: 20020924
Last Updated on STN: 20021213
Entered Medline: 20021112
AB The monoclonal IgM cold agglutinins that bind to the I/i carbohydrate Ags on the surface of RBCs all have Ig H chains encoded by the V4-34 gene segment. This mandatory use indicates that distinctive amino acid sequences may be involved in recognition. Critical amino acids exist in ***framework*** region 1 (FR1) of V4-34-encoded Ig, and these generate a specific Id determinant which apparently lies close to the I binding site. However, I binding by Id-expressing Ig can be modulated by sequences in ***complementarity*** - ***determining*** region (***CDR***)(H)3. Examination of the crystal structure of an anti-I cold agglutinin has revealed a hydrophobic patch in FR1 involving residue W7 on beta-strand A and the AVY motif (residues 23-25) on beta-strand B. In this study we used mutagenesis to show that each of the strand components of the hydrophobic patch is required for binding the I carbohydrate Ag. In addition, the crystal structure reveals that amino acids in the carboxyl-terminal region of ***CDR*** (H)3 form a surface region ***adjacent*** to the hydrophobic patch. We propose that the I carbohydrate Ag interacts simultaneously with the entire hydrophobic patch in FR1 and with the outside surface of ***CDR*** (H)3. This interaction could leave most of the conventional binding site available for binding other Ags.

L12 ANSWER 4 OF 34 MEDLINE DUPLICATE 2
ACCESSION NUMBER: 2002126474 MEDLINE
DOCUMENT NUMBER: 21851196 PubMed ID: 11861879
TITLE: Adaptive evolution of variable region genes encoding an unusual type of immunoglobulin in camelids.
AUTHOR: Su Chen; Nguyen Viet Khong; Nei Masatoshi
CORPORATE SOURCE: Institute of Molecular Evolutionary Genetics, Department of Biology, The Pennsylvania State University, Pennsylvania, USA.. su_chen@lilly.com
SOURCE: MOLECULAR BIOLOGY AND EVOLUTION, (2002 Mar) 19 (3) 205-15.
Journal code: 8501455. ISSN: 0737-4038.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200205
ENTRY DATE: Entered STN: 20020226
Last Updated on STN: 20020516
Entered Medline: 20020515
AB A ***typical*** immunoglobulin (Ig) molecule is composed of four polypeptide chains: two identical heavy (H) chains and two identical light (L) chains. This tetrameric structure is conserved in almost all jawed vertebrate species. However, it has been discovered that camels and llamas (family: Camelidae) possess a type of dimeric Ig that consists of two H chains only. These H chains do not associate with L chains, and they do not have the first constant region (CH1), which is present in the conventional Ig. In spite of these changes, the dimeric Ig maintains the normal immune function. To understand the evolution of the dimeric Ig, we studied the phylogenetic relationships of the variable region (V(H)H) genes of the dimeric Ig from Camelidae and those (V(H)) of the conventional Ig from mammals. The results showed that the V(H)H genes

form a monophyletic cluster within one of the mammalian V(H) groups, group C. We examined the type of selective force in ***complementarity*** - ***determining*** regions (CDRs) and ***framework*** regions (FRs) by comparing the rate of synonymous (dS) and nonsynonymous (dN) substitutions. We found that the results obtained from V(H)H genes were similar to those from V(H) genes in that CDRs showed an excess of dN over dS (indicating positive selection), whereas the reverse was true for FRs (purifying selection). However, when the extent of positive selection or purifying selection was investigated at each codon site, three major differences between V(H)H and V(H) genes were found. That is, very different types of selective force were observed between V(H)H and V(H) genes (1) at the sites that contact the L chain in the conventional Ig, (2) at the sites that interact with the CH1 region in the conventional Ig, and (3) in the H1 loop. Our findings suggest that adaptive evolution has occurred in the functionally important sites of the V(H)H genes to maintain the normal immune function in the dimeric Ig.

L12 ANSWER 5 OF 34 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 2002493155 MEDLINE
DOCUMENT NUMBER: 22241644 PubMed ID: 12354654
TITLE: Specific features of immunoglobulin VH genes of the Antarctic teleost Trematomus bernacchii..
AUTHOR: Oreste Umberto; Coscia Maria
CORPORATE SOURCE: Institute of Protein Biochemistry, CNR, Via Marconi, 12, 80125, Naples, Italy.. oreste@dafne.ibpe.na.cnr.it
SOURCE: GENE, (2002 Aug 7) 295 (2) 199-204.
Journal code: 7706761. ISSN: 0378-1119.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AF303534; GENBANK-AF303535; GENBANK-AF303536; GENBANK-AF303537; GENBANK-AF303538; GENBANK-AF303539; GENBANK-AF303540; GENBANK-AF303541; GENBANK-AF303542; GENBANK-AF303543; GENBANK-AF303544; GENBANK-AF303545; GENBANK-AF303546; GENBANK-AF303547; GENBANK-AF303548; GENBANK-AF303549; GENBANK-AF303550; GENBANK-AF303551; GENBANK-AF303552; GENBANK-AF303553; GENBANK-AF303554; GENBANK-AF303555; GENBANK-AF303556; GENBANK-AF303557; GENBANK-AF303558; GENBANK-AF303559; GENBANK-AF303560; GENBANK-AF303561; GENBANK-AF303562; GENBANK-AF303563; GENBANK-AF303564; GENBANK-AF303565; GENBANK-AF303566; GENBANK-AF303567; GENBANK-AF303568; GENBANK-AF303569; GENBANK-AF303570; GENBANK-AF303571; GENBANK-AF303572; GENBANK-AF303573; GENBANK-AF303574; GENBANK-AF303575
ENTRY MONTH: 200301
ENTRY DATE: Entered STN: 20021001
Last Updated on STN: 20030123
Entered Medline: 20030122

AB The somatic recombination of different germline-encoded gene segments constitutes a principal source of antibody diversity. In order to investigate the diversity in recombined gene segments encoding the immunoglobulin heavy chain of the Antarctic teleost Trematomus bernacchii, a VH library was constructed by 5'-RACE (rapid amplification of cDNA ends) using RNA isolated from the spleen of an individual specimen. Analysis of cDNA sequences of 45 rearranged VH/D/JH segments revealed specific features, such as: high number of repeats, up to 8 bp long, and palindromic sequences, especially in CDRs (complementary determining regions); occurrence of the RGYW consensus, known as mutational hot spot, higher than in other species. Sixty-four percent of single base substitutions was found within this motif. In addition, the usage of serine codons showed a clear bias for AGY in CDRs, particularly in CDR2, and for TCN in FRs (***framework*** regions). In CDRs, the frequency of non-synonymous changes was higher than that of synonymous changes. Diversity generated by insertions/deletions occurred more often than in other species; inserted bases were often repeats of ***adjacent*** bases. In particular the CDR2 showed the highest length variability as

compared to other species. Alignment of VH sequences indicated that also the gene conversion mechanism may contribute to generating diversity. These data indicate a ***CDR*** mutability higher than in other species and provide some insights into the hypermutational events that may also occur in teleosts.

L12 ANSWER 6 OF 34 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 2002175873 IN-PROCESS
DOCUMENT NUMBER: 21905407 PubMed ID: 11908722
TITLE: VH gene analysis in sporadic Burkitt's lymphoma: somatic mutation and intraclonal diversity with special reference to the tumor cells involving germinal center.
AUTHOR: Isobe Kouichi; Tamaru Jun-Ichi; Nakamura Shigeo; Harigaya Kenichi; Mikata Atsuo; Ito Hisao
CORPORATE SOURCE: Department of Radiology, Chiba University School of Medicine, Japan.
SOURCE: LEUKEMIA AND LYMPHOMA, (2002 Jan) 43 (1) 159-64. Journal code: 9007422. ISSN: 1042-8194.
PUB. COUNTRY: Switzerland
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: IN-PROCESS; NONINDEXED; Priority Journals
ENTRY DATE: Entered STN: 20020324
Last Updated on STN: 20021211
AB We analyzed the immunoglobulin heavy chain variable region (V(H)) gene in seven cases of sporadic Burkitt's lymphoma (sBL) to elucidate their cell of origin. In particular, we focused on the V(H) gene status of tumor cells involving ***adjacent*** germinal center (GC) by microdissecting histological sections. Among the seven V(H) genes V(H)1 family was found in two, V(H)3 in four, and V(H)4 in one. All rearranged V(H) genes demonstrated somatic mutations at percentages ranging from 1.4 to 7.5% (mean, 4.2%), which is a similar level to that seen in IgM-only B cells. Three out of four V(H) genes with more than 2% sequence difference from their corresponding germline counterpart showed evidence of antigen selection in their ***framework*** region 3. Three cases demonstrated signs of intraclonal diversity with a mutational frequency of 0.47-0.98%, which was 13.5-28.8 times as great as the Taq infidelity in our experimental conditions. However the level of somatic mutation and the effect of antigen selection on V(H) gene were diverse in these three cases, and the relationship between V(H) gene somatic mutation status and intraclonal diversity was unclear in sBL. In the analysis of microdissected tissues, all 20 tumor clones in the ***adjacent*** GCs showed additional replacement mutations in ***complementarity*** ***determining*** region 3, suggesting a role of antigen in tumor progression. This finding resembles the phenomenon that memory B-cells reenter into GC to undergo further affinity maturation. In contrast, 7/11 V(H) gene sequences irrelevant to GC were identical to those of the major tumor clone. Thus our findings suggested that sBL is derived from memory B-cells rather than GC B-cells, and that antigen stimulation is involved in the clonal expansion of a proportion of sBL.

L12 ANSWER 7 OF 34 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.DUPLICATE 5
ACCESSION NUMBER: 2001:348979 BIOSIS
DOCUMENT NUMBER: PREV200100348979
TITLE: Humanized immunoglobulins and methods of making the same.
AUTHOR(S): Queen, Cary L.; Selick, Harold E.
ASSIGNEE: Protein Design Labs, Inc.
PATENT INFORMATION: US 6180370 January 30, 2001
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (Jan. 30, 2001) Vol. 1242, No. 5, pp. No Pagination. e-file.
ISSN: 0098-1133.
DOCUMENT TYPE: Patent
LANGUAGE: English
AB Novel methods for producing, and compositions of, humanized immunoglobulins having one or more ***complementarity***

determining regions (***CDR*** 's) and possible additional amino acids from a donor immunoglobulin and a ***framework*** region from an accepting human immunoglobulin are provided. Each humanized immunoglobulin chain will usually comprise, in addition to the ***CDR*** 's, amino acids from the donor immunoglobulin ***framework*** that are, e.g., capable of interacting with the ***CDR*** 's to effect binding affinity, such as one or more amino acids which are immediately ***adjacent*** to a ***CDR*** in the donor immunoglobulin or those within about about 3 ANG as predicted by molecular modeling. The heavy and light chains may each be designed by using any one or all of various position criteria. When combined into an intact antibody, the humanized immunoglobulins of the present invention will be substantially non-immunogenic in humans and retain substantially the same affinity as the donor immunoglobulin to the antigen, such as a protein or other compound containing an epitope.

L12 ANSWER 8 OF 34 MEDLINE DUPLICATE 6
ACCESSION NUMBER: 2001213243 MEDLINE
DOCUMENT NUMBER: 21103249 PubMed ID: 11160277
TITLE: Novel secondary Ig VH gene rearrangement and in-frame Ig heavy chain ***complementarity*** - ***determining*** region III insertion/deletion variants in de novo follicular lymphoma.
AUTHOR: Kobrin C; Bendandi M; Kwak L
CORPORATE SOURCE: Intramural Research Support Program, Science Applications International Corp.-Frederick, National Cancer Institute-Frederick Cancer Research and Development Center, Frederick, MD 21702, USA.
CONTRACT NUMBER: NO1-CO-56000 (NCI)
SOURCE: JOURNAL OF IMMUNOLOGY, (2001 Feb 15) 166 (4) 2235-43. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200104
ENTRY DATE: Entered STN: 20010425
Last Updated on STN: 20010425
Entered Medline: 20010419
AB Human germinal center B cell tumors retain the ability of their nontransformed counterparts to somatically hypermutate Ig V genes by nucleotide substitution. Among a survey of 60 primary previously untreated, clonal, follicular lymphomas we have identified a ***rare*** V(H) rearrangement variant and two other in-frame nucleotide insertion/deletion variants within ***complementarity*** - ***determining*** region III of the Ig heavy chain. The neoplastic origin of the V(H) rearrangement variant was directly demonstrated in cells isolated by microdissection from malignant follicles. In all three cases a common clonal origin for the variants was demonstrated by ***complementarity*** - ***determining*** region III nucleotide sequence homology and shared somatic mutations in germline encoded positions in ***framework*** region IV. The monoclonal nature of the tumors was independently confirmed by demonstrating a single t(14;18) translocation breakpoint in the two cases with a detectable translocation. All the variants occurred in functional V(H) rearrangements, which in two cases were directly shown to encode functional Ab molecules. Both recombination-activating genes 1 and 2 were expressed in lymph node tumor cells containing the V(H) rearrangement variant, although recombination-activating gene expression among a panel of lymphomas was not limited to this variant.

L12 ANSWER 9 OF 34 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 2002:261483 BIOSIS
DOCUMENT NUMBER: PREV200200261483
TITLE: Molecular histogenesis of post-transplant lymphoproliferative disorders.
AUTHOR(S): Capello, Daniela (1); Vivenza, Daniela (1); Muti, Giuliana;

Rossi, Davide (1); Berra, Eva (1); Franceschetti, Silvia (1); Oreste, Pierluigi; Viglio, Alessandra; Morra, Enrica; Paulli, Marco; Gaidano, Gianluca (1)
CORPORATE SOURCE: (1) Department of Medical Sciences, Amedeo Avogadro University of Eastern Piedmont, Novara Italy
SOURCE: Blood, (November 16, 2001) Vol. 98, No. 11 Part 1, pp. 805a-806a. http://www.bloodjournal.org/. print.
Meeting Info.: 43rd Annual Meeting of the American Society of Hematology, Part 1 Orlando, Florida, USA December 07-11, 2001
ISSN: 0006-4971.
DOCUMENT TYPE: Conference
LANGUAGE: English
AB Post-transplant lymphoproliferative disorders (PTLD) represent a heterogeneous group of lymphoproliferations arising in transplant recipients. PTLD comprise a histologic spectrum ranging from polyclonal hyperplasia to frank lymphoma or multiple myeloma. The histogenetic derivation of PTLD has not been addressed in detail. In order to refine the pathogenesis and histogenesis of the disease, we investigated the usage, the mutation pattern and the antigen selection process of immunoglobulin variable (IgV) heavy (H) and light (L) chain genes in a panel of 24 PTLD arising in solid organ transplant recipients. In parallel; other genotypic and phenotypic markers of histogenesis were investigated and correlated to IgV mutations. These included BCL-6 and CD138 expression, which segregate the germinal center (GC) stage of B-cell differentiation (BCL-6+/CD138-) from later stages of maturation (BCL-6-/CD138+); and BCL-6 mutations, which are accumulated during GC transit of B-cells. Evidence of antigen selection of IgV genes was analyzed by the binomial (Chang-Casali) and the multinomial statistical methods. PTLD utilized all most common IgV genes, with no specific bias for a given IgVH or IgVL family. Twenty of 22 (91%) PTLD were found to carry somatic mutations in IgVH and/or IgVL genes, at a frequency ranging from 2.80% to 24.7%. Notably, both IgVH and IgVL genes of two large B-cell immunoblastic lymphomas harbored crippling mutations, i.e. stop codon mutations within originally functional rearrangements, suggesting a cellular origin from pre-apoptotic GC B-cells that have lost the ability to express antigen receptors. By combining the IgVH and IgVL chain analysis, evidence for a significant counterselection of replacement (R) mutations within the IgV ***framework*** regions (FR) was observed in 12/17 (70%) PTLD, while evidence of antigen selection, signified by clustering of R mutations in the complementary determining regions (***CDR***), was observed in 8/17 (47%) cases. The BCL-6 gene harbored somatic mutations in 7/23 (30%) cases. The frequency of mutation in each individual case ranged from 0.07 to 1.39X10-3/bp. All PTLD mutated in the BCL-6 gene were also mutated in IgV genes. With respect to phenotypic markers, virtually all PTLD tested failed to express BCL-6 and CD138. The BCL-6-/CD138- phenotype of PTLD is at variance with that of other GC-related lymphomas arising in the setting of immunodeficiency, namely AIDS-related lymphomas, which display either a ***typical*** GC-related phenotype (BCL-6+/CD138-) or a classical post-GC phenotype (BCL-6-/CD138+). The implications of these data are multifold. First, virtually all PTLD derive from GC-related B-cells, independent of their differentiation stage, morpho-phenotypic profile and site of origin. Second, the combination of IgV and BCL-6 mutations coupled to absent expression of both BCL-6 and CD138 suggests that PTLD conceivably reflect a post-GC stage of B-cell differentiation which has not yet undergone preterminal maturation. Third, the counterselection of R mutations within the FR regions suggests that most PTLD originate from mutating B-cell clones selected, at least for some period of time, for expression of functional antigen receptors. Finally, the preferential clustering of R mutations in ***CDR*** regions indicate that a fraction of PTLD derive from antigen-experienced B-cells.

L12 ANSWER 10 OF 34 MEDLINE DUPLICATE 7
ACCESSION NUMBER: 2001357851 MEDLINE
DOCUMENT NUMBER: 21311885 PubMed ID: 11418663
TITLE: Structural effects of ***framework*** mutations on a

humanized anti-lysozyme antibody.
AUTHOR: Holmes M A; Buss T N; Foote J
CORPORATE SOURCE: Program in Molecular Medicine, Fred Hutchinson Cancer Research Center, Seattle, WA 98109, USA.
SOURCE: JOURNAL OF IMMUNOLOGY, (2001 Jul 1) 167 (1) 296-301. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
OTHER SOURCE: PDB-H27S; PDB-H71A; PDB-H71R; PDB-H71V
ENTRY MONTH: 200109
ENTRY DATE: Entered STN: 20010924
Last Updated on STN: 20010924
Entered Medline: 20010920
AB A humanized version of the mouse anti-lysozyme Ab D1.3 was previously constructed as an Fv fragment and its structure was crystallographically determined in the free form and in complex with lysozyme. Here we report five new crystal structures of single-amino acid substitution mutants of the humanized Fv fragment, four of which were determined as Fv-lysozyme complexes. The crystals were isomorphous with the parent forms, and were refined to free R values of 28-31% at resolutions of 2.7-2.9 A. Residue 27 in other Abs has been implicated in stabilizing the conformation of the first ***complementarity*** - ***determining*** region (***CDR***) of the H chain, residues 31-35. We find that a Phe-to-Ser mutation at 27 alters the conformation of immediately ***adjacent*** residues, but this change is only weakly transmitted to Ag binding residues in the nearby ***CDR*** . Residue 71 of the H chain has been proposed to control the relative disposition of H chain CDRs 1 and 2, based on the bulk of its side chain. However, in structures we determined with Val, Ala, or Arg substituted in place of Lys at position 71, no significant change in the conformation of CDRs 1 and 2 was observed.

L12 ANSWER 11 OF 34 MEDLINE DUPLICATE 8
ACCESSION NUMBER: 2001107058 MEDLINE
DOCUMENT NUMBER: 21034368 PubMed ID: 11193052
TITLE: Prediction of the interacting surfaces in a trimolecular complex formed between the major dust mite allergen Der p 1, a mouse monoclonal anti-Der p 1 antibody, and its anti-idiotype.
AUTHOR: Furtado P B; Furmonaviciene R; McElveen J; Sewell H F; Shakib F
CORPORATE SOURCE: Division of Molecular and Clinical Immunology, University of Nottingham, Faculty of Medicine and Health Sciences, Nottingham NG7 2UH, UK.
SOURCE: MOLECULAR PATHOLOGY, (2000 Dec) 53 (6) 324-32. Journal code: 9706282. ISSN: 1366-8714.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200102
ENTRY DATE: Entered STN: 20010322
Last Updated on STN: 20010322
Entered Medline: 20010208
AB BACKGROUND: Two mouse monoclonal antibodies (mAbs) have been described recently; namely, mAb 2C7 (IgG2b kappa), which is directed against the major house dust mite allergen Der p 1, and mAb 2G10 (IgG1 kappa), which is an anti-idiotypic antibody raised against mAb 2C7. The anti-idiotype mAb 2G10 does not block the binding of mAb 2C7 to Der p 1, which means that mAb 2C7 can simultaneously bind to Der p 1 and to mAb 2G10, thereby generating a trimolecular complex consisting of antigen-idiotype-anti-idiotype. AIMS: To sequence and model the V region of the anti-idiotypic antibody mAb 2G10 to enable the prediction of the interacting surfaces in the trimolecular complex consisting of Der p 1-mAb 2C7-mAb 2G10. METHODS: DNA sequencing of mAb 2G10 was carried out and the Swiss Model and Swiss PDB-Viewer programs were used to build a three dimensional model of the

trimolecular complex. RESULTS: Complementarity of shape and charge was revealed when comparing the protrusion of the previously determined Der p 1 epitope (Leu147-Gln160) with the cavity formed by the ***complementarity*** ***determining*** regions (CDRs) of mAb 2C7. Such complementarity was also observed between the mAb 2C7 epitope predicted to be recognised by mAb 2G10 (residues Lys19 from ***framework*** region 1 (FRW1) and Ser74-Gln81 from FRW3) and residues from the CDRs of mAb 2G10 (a negatively charged patch flanked by the residues Asp55H/Glu58H and Glu27L/Glu27cL). As expected, the location of the mAb 2C7 epitope recognised by mAb 2G10 does not appear to interfere with the binding of Der p 1 to mAb 2C7. CONCLUSION: Although the results obtained represent only an approximation, they nevertheless provide a ***rare*** insight into how an antigen (Der p 1) might bind to its antibody (mAb 2C7) while in complex with an anti-idiotype (mAb 2G10).

L12 ANSWER 12 OF 34 MEDLINE DUPLICATE 9
ACCESSION NUMBER: 2000010049 MEDLINE
DOCUMENT NUMBER: 20010049 PubMed ID: 10540323
TITLE: Targeting and subsequent selection of somatic hypermutations in the human V kappa repertoire.
AUTHOR: Foster S J; Dorner T; Lipsky P E
CORPORATE SOURCE: Department of Internal Medicine, Harold C. Simmons Arthritis Research Center, University of Texas Southwestern Medical Center, Dallas 75235-8884, USA.
CONTRACT NUMBER: AI3 1229 (NIAID)
SOURCE: EUROPEAN JOURNAL OF IMMUNOLOGY, (1999 Oct) 29 (10) 3122-32. Journal code: 1273201. ISSN: 0014-2980.
PUB. COUNTRY: GERMANY: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199911
ENTRY DATE: Entered STN: 20000111
Last Updated on STN: 20000111
Entered Medline: 19991116
AB The number and distribution of nucleotide substitutions in human VkappaJkappa genes were examined using a PCR technique that analyzed nonproductive and productive rearrangements amplified from genomic DNA of individual B cells. The results indicate that the mutational mechanism introduces replacement (R) mutations comparably throughout the length of the VkappaJkappa rearrangement, but tends to target specific triplets. Moreover, hotspots of mutational activity were identified in ***complementarity*** ***determining*** regions (***CDR***). A marked increase in the frequency of R mutations in ***CDR*** was noted when productive were compared to nonproductive rearrangements, indicating that these were selected into the expressed repertoire. Of note, amino acids encoded by codons ***adjacent*** to hotspots of mutation were also positively selected implying that similar regions were targeted for hypermutation and subsequent selection. In contrast to the distribution of ***CDR*** mutations, R mutations in the ***framework*** (FR) regions tended to be eliminated from productive VkappaJkappa rearrangements, implying that the somatic hypermutational machinery frequently introduced amino acid changes that were deleterious to the structural integrity of the kappa chain protein. The difference in the ratio of R to silent mutations in ***CDR*** and FR in the expressed repertoire, therefore, reflects the summation of positive selection of R mutations in the ***CDR*** and the elimination of R mutations in the FR. The data indicate that the balance between targeted mutation of VkappaJkappa rearrangements and subsequent selection and elimination governs the pattern of mutations manifest within the expressed kappa repertoire.

L12 ANSWER 13 OF 34 EMBASE COPYRIGHT 2003 ELSEVIER SCI. B.V.DUPLICATE 10
ACCESSION NUMBER: 1999126949 EMBASE
TITLE: Structural mimicry of canonical conformations in antibody ***hypervariable*** loops using cyclic peptides containing a heterochiral diproline template.

AUTHOR: Favre M.; Moehle K.; Jiang L.; Pfeiffer B.; Robinson J.A.
CORPORATE SOURCE: J.A. Robinson, Institute of Organic Chemistry, University of Zurich, Winterthurerstrasse 190, 8057 Zurich, Switzerland. robinson@oci.unizh.ch
SOURCE: Journal of the American Chemical Society, (31 Mar 1999) 121/12 (2679-2685).
ISSN: 0002-7863 CODEN: JACSAT
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Analyses of high resolution crystal structures have shown that antibody ***hypervariable*** loops L1, L2, and L3 from the light chain, as well as H1 and H2 from the heavy chain, can be assigned to only a small family of canonical conformations. We describe here attempts to generate structural mimetics of L2, L3, and H2 canonical conformations, which are .beta.-hairpin structures connecting ***adjacent*** antiparallel .beta.-strands. The five mimetics studied comprise cyclic peptides, in which the ***CDR*** loop has been transplanted from the immunoglobulin ***framework*** onto a D-Pro-L-Pro template. Their preferred conformations have been studied by NMR and MD with time-averaged, NOE-derived distance restraints. The results show that accurate mimetics of L3 and H2 loops can be obtained, whereas the L2 canonical conformation, which appears to be inherently strained, could not be mimicked in this way. For example, an eight-residue L3 loop from antibody HC19 attached to the D-Pro- L-Pro template adopts not only a backbone hairpin conformation but also aromatic-aromatic T-stacking interactions between tryptophan side-chains, that are essentially identical to those in the antibody crystal structure. This straightforward and effective approach to hairpin design may be of great value for generating small molecule mimetics of hairpin loops on proteins of diverse function.

L12 ANSWER 14 OF 34 MEDLINE DUPLICATE 11
ACCESSION NUMBER: 1999453805 MEDLINE
DOCUMENT NUMBER: 99453805 PubMed ID: 10524280
TITLE: Tertiary structure of human lambda 6 light chains.
AUTHOR: Pokkuluri P R; Solomon A; Weiss D T; Stevens F J; Schiffer M
CONTRACT NUMBER: CA10056 (NCI)
DK43757 (NIDDK)
SOURCE: AMYLOID, (1999 Sep) 6 (3) 165-71.
Journal code: 9433802. ISSN: 1350-6129.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199911
ENTRY DATE: Entered STN: 20000111
Last Updated on STN: 20000111
Entered Medline: 19991116
AB AL amyloidosis is a disease process characterized by the pathologic deposition of monoclonal light chains in tissue. To date, only limited information has been obtained on the molecular features that render such light chains amyloidogenic. Although protein products of the major human V kappa and V lambda gene families have been identified in AL deposits, one particular subgroup--lambda 6--has been found to be preferentially associated with this disease. Notably, the variable region of lambda 6 proteins (V lambda 6) has distinctive primary structural features including the presence in the third ***framework*** region (FR3) of two additional amino acid residues that distinguish members of this subgroup from other types of light chains. However, the structural consequences of these alterations have not been elucidated. To determine if lambda 6 proteins possess unique tertiary structural features, as compared to light chains of other V lambda subgroups, we have obtained x-ray diffraction data on crystals prepared from two recombinant V lambda

6 molecules. These components, isolated from a bacterial expression system, were generated from lambda 6-related cDNAs cloned from bone marrow-derived plasma cells from a patient (Wil) who had documented AL amyloidosis and another (Jto) with multiple myeloma and tubular cast nephropathy, but no evident fibrillar deposits. The x-ray crystallographic analyses revealed that the two-residue insertion located between positions 68 and 69 (not between 66 and 67 as previously surmised) extended an existing loop region that effectively increased the surface area ***adjacent*** to the first ***complementarity*** ***determining*** region (CDR1). Further, an unusual interaction between the Arg 25 and Phe 2 residues commonly found in lambda 6 molecules was noted. However, the structures of V lambda 6 Wil and Jto also differed from each other, as evidenced by the presence in the latter of certain ionic and hydrophobic interactions that we posit increased protein stability and thus prevented amyloid formation.

L12 ANSWER 15 OF 34 MEDLINE DUPLICATE 12
ACCESSION NUMBER: 1999326368 MEDLINE
DOCUMENT NUMBER: 99326368 PubMed ID: 10398393
TITLE: Comparison of the three-dimensional structures of a humanized and a chimeric Fab of an anti-gamma-interferon antibody.
AUTHOR: Fan Z C; Shan L; Goldsteen B Z; Guddat L W; Thakur A; Landolfi N F; Co M S; Vasquez M; Queen C; Ramsland P A; Edmundson A B
CORPORATE SOURCE: Oklahoma Medical Research Foundation, 825 NE 13th Street, Oklahoma City, OK 73104, USA.
CONTRACT NUMBER: CA 72803 (NCI)
SOURCE: JOURNAL OF MOLECULAR RECOGNITION, (1999 Jan-Feb) 12 (1) 19-32. Ref: 78
Journal code: 9004580. ISSN: 0952-3499.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200003
ENTRY DATE: Entered STN: 20000320
Last Updated on STN: 20000320
Entered Medline: 20000309

AB The objective of this work is to compare the three-dimensional structures of "humanized" and mouse-human chimeric forms of a murine monoclonal antibody elicited against human gamma-interferon. It is also to provide structural explanations for the small differences in the affinities and biological interactions of the two molecules for this antigen. Antigen-binding fragments (Fabs) were produced by papain hydrolysis of the antibodies and crystallized with polyethylene glycol (PEG) 8,000 by nearly identical microseeding procedures. Their structures were solved by X-ray analyses at 2.9 A resolution, using molecular replacement methods and crystallographic refinement. Comparison of these structures revealed marked similarities in the light (L) chains and near identities of the constant (C) domains of the heavy (H) chains. However, the variable (V) domains of the heavy chains exhibited substantial differences in the conformations of all three ***complementarity*** - ***determining*** regions (CDRs), and in their first ***framework*** segments (FR1). In FR1 of the humanized VH, the substitution of serine for proline in position 7 allowed the N-terminal segment (designated strand 4-1) to be closely juxtaposed to an ***adjacent*** strand (4-2) and form hydrogen bonds ***typical*** of an antiparallel beta-pleated sheet. The tightening of the humanized structure was relayed in such a way as to decrease the space available for the last portion of HFR1 and the first part of HCDR1. This compression led to the formation of an alpha-helix involving residues 25-32. With fewer steric constraints, the corresponding segment in the chimeric Fab lengthened by at least 1 A to a random coil which terminated in a single turn of 310 helix. In the humanized Fab, HCDR1, which is sandwiched between HCDR2 and HCDR3,

significantly influenced the structures of both regions. HCDR2 was forced into a bent and twisted orientation different from that in the chimeric Fab, both at the crown of the loop (around proline H52a) and at its base. As in HCDR1, the last few residues of HCDR2 in the humanized Fab were compressed into a space-saving alpha-helix, contrasting with a more extended 310 helix in the chimeric form. HCDR3 in the humanized Fab was also adjusted in shape and topography. The observed similarities in the functional binding activities of the two molecules can be rationalized by limited induced fit adjustments in their structures on antigen binding. While not perfect replicas, the two structures are testimonials to the progress in making high affinity monoclonal antibodies safe for human use.

L12 ANSWER 16 OF 34 MEDLINE DUPLICATE 13
ACCESSION NUMBER: 1998414289 MEDLINE
DOCUMENT NUMBER: 98414289 PubMed ID: 9743357
TITLE: Molecular modeling of an anti-DNA autoantibody (V-88) and mapping of its V region epitopes recognized by heterologous and autoimmune antibodies.
AUTHOR: Hobby P; Ward F J; Denbury A N; Williams D G; Staines N A; Sutton B J
CORPORATE SOURCE: The Randall Institute, Biomedical Sciences Division, King's College London, United Kingdom.
SOURCE: JOURNAL OF IMMUNOLOGY, (1998 Sep 15) 161 (6) 2944-52. Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199810
ENTRY DATE: Entered STN: 19981020
Last Updated on STN: 19981020
Entered Medline: 19981006

AB Anti-DNA autoantibodies are a characteristic feature of human systemic lupus erythematosus (SLE) and lupus diseases in the mouse. V-88 is an IgG1/kappa ssDNA-binding Ab, derived from a lupus mouse, that bears a cross-species, cross-reactive Id (CRI) that has been implicated in the pathogenesis of both human and murine disease. A linear epitope map of V-88 has been determined with anti-idiotypic antisera obtained from rabbits, and candidate sequences for the idiotopes of the CRI have been proposed. We now report the modeling of the three-dimensional structure of the V regions of Ab V-88, to map the location of these idiotopes. The V region ***framework*** structure was derived from those of crystallographically determined Ab structures, and the ***complementarity*** ***determining*** region (***CDR***) structures were based upon the set of canonical structures adopted by these loop regions in Abs of known structure. One of the idiotopes is an extensive, highly accessible epitope consisting of ***framework*** regions spatially ***adjacent*** to CDR2 in the heavy chain. Epitopes recognized by an anti-idiotypic rabbit antiserum were compared with those recognized by autoimmune sera from SLE-prone mice, and common features were identified. By analogy with the crystal structure of an anti-DNA Ab BV04-01 complexed with a trinucleotide, the modeled structure also suggests a mode of binding of ssDNA to V-88. The location of the candidate CRI, although within the ***framework*** region of VH, is such that it could influence Ag specificity.

L12 ANSWER 17 OF 34 MEDLINE DUPLICATE 14
ACCESSION NUMBER: 1998395165 MEDLINE
DOCUMENT NUMBER: 98395165 PubMed ID: 9727062
TITLE: Antigen-driven clonal proliferation of B cells within the target tissue of an autoimmune disease. The salivary glands of patients with Sjogren's syndrome.
AUTHOR: Stott D I; Hiepe F; Hummel M; Steinhauser G; Berek C
CORPORATE SOURCE: University Department of Immunology, Western Infirmary, Glasgow G11 6NT, Scotland, United Kingdom..
d.i.stott@clinmed.gla.ac.uk

SOURCE: JOURNAL OF CLINICAL INVESTIGATION, (1998 Sep 1) 102 (5) 938-46.
Journal code: 7802877. ISSN: 0021-9738.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199810
ENTRY DATE: Entered STN: 19981020
Last Updated on STN: 19981020
Entered Medline: 19981006

AB Structures resembling germinal centers are seen in the salivary glands of patients with Sjogren's syndrome, but it is not known whether the microenvironment of these cell clusters is sufficient for the induction of a germinal center response. Therefore, we cloned and sequenced rearranged Ig V genes expressed by B cells isolated from sections of labial salivary gland biopsies from two Sjogren's syndrome patients. Rearranged V genes from B cells within one cell cluster were polyclonal and most had few somatic mutations. Two ***adjacent*** clusters from another patient each contained one dominant B cell clone expressing hypermutated V genes. None of the rearranged V genes was found in both clusters, suggesting that cells are unable to migrate out into the surrounding tissue and seed new clusters. The ratios of replacement to silent mutations in the ***framework*** and ***complementarity*** ***determining*** regions suggest antigen selection of high-affinity mutants. These results show that an antigen-driven, germinal center-type B cell response is taking place within the salivary glands of Sjogren's syndrome patients. In view of the recent demonstration of a germinal center response within the rheumatoid synovial membrane and the existence of similar structures in the target tissues of other autoimmune diseases, we propose that germinal center- type responses can be induced in the nonlymphoid target tissues of a variety of autoimmune diseases.

L12 ANSWER 18 OF 34 MEDLINE DUPLICATE 15

ACCESSION NUMBER: 1999030463 MEDLINE
DOCUMENT NUMBER: 99030463 PubMed ID: 9811544

TITLE: Crystal structures of a rat anti-CD52 (CAMPATH-1) therapeutic antibody Fab fragment and its humanized counterpart.

AUTHOR: Cheetham G M; Hale G; Waldmann H; Bloomer A C
CORPORATE SOURCE: MRC Laboratory of Molecular Biology, Hills Road, Cambridge, CB2 2QH, UK.

SOURCE: JOURNAL OF MOLECULAR BIOLOGY, (1998 Nov 20) 284 (1) 85-99.
Journal code: 2985088R. ISSN: 0022-2836.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: PDB-1RBEY; PDB-R1BEYST
ENTRY MONTH: 199902
ENTRY DATE: Entered STN: 19990311
Last Updated on STN: 20030313
Entered Medline: 19990225

AB The CAMPATH-1 family of antibodies are able systematically to lyse human lymphocytes with human complement by targeting the small cell-surface glycoprotein CD52, commonly called the CAMPATH-1 antigen. These antibodies have been used clinically for several years, providing therapy for patients with a variety of immunologically mediated diseases. We report here the first X-ray crystallographic analyses of a Fab fragment from a rat antibody, the original therapeutic monoclonal CAMPATH-1G and its humanized counterpart CAMPATH-1H, into which the six ***complementarity*** - ***determining*** regions of the rat antibody have been introduced. These structures have been refined at 2.6 A and 3.25 A resolution, respectively. The VL domains of ***adjacent*** molecules of CAMPATH-1H form a symmetric dimer within the crystals with an inter-molecular extended beta-sheet as seen in light chain dimers of the kappa class. Crystals of CAMPATH-1G have translational pseudo-symmetry.

Within the antibody-combining sites, which are dominated by the protrusion of LysH52b and LysH53 from ***hypervariable*** loop H2, the charge distribution and overall integrity are highly conserved, but large changes in the position of loop H1 are observed and an altered conformation of loop H2. The major determinants of this are ***framework*** residues H71 and H24, whose identity differs in these two antibodies. These structures provide a detailed structural insight into the transplantation of an intact antibody-combining site between a rodent and a human ***framework*** , and provide an increased understanding of the specificity and antigen affinity of this pair of CAMPATH-1 antibodies for CD52. This study forms the structural basis for future modification and design of more effective antibodies to this important antigen.

L12 ANSWER 19 OF 34 MEDLINE DUPLICATE 16
ACCESSION NUMBER: 97360156 MEDLINE
DOCUMENT NUMBER: 97360156 PubMed ID: 9217184
TITLE: Immunoglobulin heavy chain diversity genes rearrangement pattern indicates that MALT-type gastric lymphoma B cells have undergone an antigen selection process.
AUTHOR: Bertoni F; Cazzaniga G; Bosshard G; Roggero E; Barbazza R; De Boni M; Capella C; Pedrinis E; Cavalli F; Biondi A; Zucca E
CORPORATE SOURCE: Servizio Oncologico Cantonale, Ospedale San Giovanni, Bellinzona, Switzerland.
SOURCE: BRITISH JOURNAL OF HAEMATOLOGY, (1997 Jun) 97 (4) 830-6. Journal code: 0372544. ISSN: 0007-1048.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-AF016198; GENBANK-AF016199; GENBANK-AF016200; GENBANK-AF016201; GENBANK-AF016202; GENBANK-AF016203; GENBANK-AF016204; GENBANK-AF016205; GENBANK-AF016206; GENBANK-AF016207; GENBANK-AF016208; GENBANK-AF016209; GENBANK-AF016210; GENBANK-AF016211; GENBANK-AF016212; GENBANK-AF016213; GENBANK-AF016214; GENBANK-AF016215; GENBANK-AF016216
ENTRY MONTH: 199707
ENTRY DATE: Entered STN: 19970805
Last Updated on STN: 20000303
Entered Medline: 19970723

AB Gastric MALT lymphoma usually develops from chronic gastritis, the vast majority of which (>90%) is associated with Helicobacter pylori infection. We sequenced the third ***complementarity*** ***determining*** region (CDR3) of immunoglobulin heavy chain genes in 19 gastric MALT lymphoma clones to determine the pattern of variable (V), diversity (D) and joining (J) gene utilization during immunoglobulin gene rearrangement. DNA was extracted from paraffin-embedded sections and the rearranged CDR3 regions were amplified using a semi-nested polymerase chain reaction (with primers complementary to the conserved ***framework*** -three segment of the variable region [FR3A] and J regions). The DNA used for cloning and sequencing was obtained after purification of monoclonal bands excised from polyacrylamide gels. The N-D-N region specific to each clone was compared with known germline D sequences. Similarly to that observed in normal and leukaemic B cells, our series of gastric MALT lymphomas showed apparent preferential utilization of genes from the DXP family. In two cases no similarity between the CDR3 nucleotide sequences of the neoplastic clones and the known germline D sequences could be found. In 10/19 analysed alleles the lymphoma B-cell clones appeared to contain two D gene segments (D-D recombination), a ***rare*** occurrence in normal individuals but one which has been described as a significant event in the determination of idiotype expression and antigen-binding affinity. Remarkably, despite the use of different D and J segments, the resultant amino acid sequences matched in two patients, suggesting the presence of a common selecting antigen. The observed pattern of D gene rearrangement suggests that MALT lymphoma B-cell clones have undergone antigen

selection, which seems to indicate that the antigen stimulation plays a pivotal role in the development of the lymphoma.

L12 ANSWER 20 OF 34 MEDLINE DUPLICATE 17
ACCESSION NUMBER: 1998147062 MEDLINE
DOCUMENT NUMBER: 98147062 PubMed ID: 9486108
TITLE: Natural antibodies that react with V-region peptide epitopes of DNA-binding antibodies are made by mice with systemic lupus erythematosus as disease develops.
AUTHOR: Ward F J; Knies J E; Cunningham C; Harris W J; Staines N A
CORPORATE SOURCE: Infection and Immunity Research Group, King's College London, UK.
SOURCE: IMMUNOLOGY, (1997 Nov) 92 (3) 354-61.
Journal code: 0374672. ISSN: 0019-2805.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199803
ENTRY DATE: Entered STN: 19980319
Last Updated on STN: 19980319
Entered Medline: 19980309
AB Cross-reactive idiotypes (CRI) have been detected on anti-DNA autoantibodies associated with lesions ***typical*** of systemic lupus erythematosus. In order to analyse the antigenic make up of idiotypes on anti-DNA monoclonal antibodies (mAb) V-88 (IgG1 kappa) and F-423 (IgG3 kappa), derived respectively from an adult (NZB x NZW)F1 and a fetal MRL/Mp-lpr/lpr mouse, a set of overlapping hexapeptides representing the VH and VL regions of mAb V-88 and F-423 were synthesized and reacted with a range of sera in pepscan enzyme-linked immunosorbent assays (ELISA) taken from normal and lupus mouse strains. Serum pools were collected both from normal BALB/c and lupus MRL/Mp-lpr/lpr and (NZB x NZW)F1 mice at 10, 20 and 30 weeks of age and analysed for the presence of spontaneously produced anti-V-region peptide IgM and IgG antibodies. IgM antibodies from both the lupus mice reacted with the same V-region epitopes, and although some epitopes mapped to similar locations in the two mAb, the maps for V-88 and F-423 were not identical. In MRL/Mp-lpr/lpr mice, as lupus disease progressed there was a switch from IgM antibodies to IgG anti-peptide antibodies whose specificity for the peptide antigens coincided with but was better defined than that of the IgM antibodies. The identified idiotopes were located in both complementary determining regions (***CDR***) and ***framework*** region (FR) regions, indicating that some contribute to CRI shared by other related antibodies, while others were unique to either mAb V-88 or F-423. In conclusion, we have dissected and identified a mosaic of antibody V-region idiotopes that contribute to the idiotype of an anti-DNA autoantibody and against which autoantibodies are made naturally in lupus disease.

L12 ANSWER 21 OF 34 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1996:447099 CAPLUS
DOCUMENT NUMBER: 125:140550
TITLE: Humanized immunoglobulins
INVENTOR(S): Queen, Cary L.; Selick, Harold E.
PATENT ASSIGNEE(S): Protein Design Labs, Inc., USA
SOURCE: U.S., 139 pp., Cont.-in-part of U.S. Ser. No. 590,274, abandoned.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 4
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 5530101	A	19960625	US 1990-634278	19901219
ZA 8909956	A	19901031	ZA 1989-9956	19891228
EP 682040	A1	19951115	EP 1995-105609	19891228

EP 682040 B1 19990825
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE
JP 11004694 A2 19990112 JP 1998-4334 19891228
EP 939127 A2 19990901 EP 1998-204240 19891228
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE
CA 2328851 C 20020813 CA 1989-2328851 19891228
DD 296964 A5 19911219 DD 1990-337159 19900117
CA 2098404 AA 19920620 CA 1991-2098404 19911219
CA 2098404 C 20020820
WO 9211018 A1 19920709 WO 1991-US9711 19911219
W: AT, AU, BB, BG, BR, CA, CH, CS, DE, DK, ES, FI, GB, HU, JP, KP, KR, LK, LU, MG, MN, MW, NL, NO, PL, RO, SD, SE, SU
RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GN, GR, IT, LU, MC, ML, MR, NL, SE, SN, TD, TG
AU 9191726 A1 19920722 AU 1991-91726 19911219
AU 671949 B2 19960919
EP 566647 A1 19931027 EP 1992-903551 19911219
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, MC, NL, SE
JP 06503963 T2 19940512 JP 1991-503758 19911219
JP 3276369 B2 20020422 JP 1992-503758 19911219
US 5585089 A 19961217 US 1995-477728 19950607
US 5693761 A 19971202 US 1995-474040 19950607
US 5693762 A 19971202 US 1995-487200 19950607
US 6180370 B1 20010130 US 1995-484537 19950607
SG 78258 A1 20010220 SG 1996-7855 19960413
AU 9675481 A1 19970220 AU 1996-75481 19961219
PRIORITY APPLN. INFO.: US 1988-290975 B2 19881228
US 1989-310252 B2 19890213
US 1990-590274 B2 19900928
CA 1989-2006865 A3 19891228
EP 1990-903576 A3 19891228
EP 1995-105609 A3 19891228
JP 1990-503677 A3 19891228
US 1990-634278 A 19901219
WO 1991-US9711 A 19911219

AB Novel methods for producing, and compns. of, humanized Igs having one or more complementarity detg. regions (***CDR*** 's) and possible addnl. amino acids from a donor Ig and a . ***framework*** region from an accepting human Ig are provided. Each humanized Ig chain will usually comprise, in addn. to the ***CDR*** 's, amino acids from the donor Ig ***framework*** that are, e.g., capable of interacting with the ***CDR*** 's to effect binding affinity, such as one or more amino acids which are immediately ***adjacent*** to a ***CDR*** in the donor Ig or those within about 3 .ANG. as predicted by mol. modeling. The heavy and light chains may each be designed by using any one or all of various position criteria. When combined into an intact antibody, the humanized Igs of the present invention will be substantially non-immunogenic in humans and retain substantially the same affinity as the donor Ig to the antigen, such as a protein or other compd. contg. an epitope. In example, prepd. were humanized antibodies specific for Tac (activated T cell), gB and gD glycoproteins of herpes simplex virus, CD33 antigen, p75 of interleukin 2 receptor, gH glycoprotein of cytomegalovirus.

L12 ANSWER 22 OF 34 MEDLINE DUPLICATE 18
ACCESSION NUMBER: 96208119 MEDLINE
DOCUMENT NUMBER: 96208119 PubMed ID: 8623912
TITLE: Light chain cardiomyopathy. Structural analysis of the light chain tissue deposits.
COMMENT: Comment in: Am J Pathol. 1996 May;148(5):1339-44
AUTHOR: Gallo G; Goni F; Boctor F; Vidal R; Kumar A; Stevens F J; Frangione B; Ghiso J
CORPORATE SOURCE: Department of Pathology, New York University Medical Center, New York 10016, USA.
CONTRACT NUMBER: AR02594 (NIAMS)
DK43757 (NIDDK)
SOURCE: AMERICAN JOURNAL OF PATHOLOGY, (1996 May) 148 (5) 1397-406.
Journal code: 0370502. ISSN: 0002-9440.

PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199606
ENTRY DATE: Entered STN: 19960627
Last Updated on STN: 19960627
Entered Medline: 19960620

AB Cardiomyopathy due to monoclonal light chain deposits is a complication of plasma cell disorders. The deposits may be either fibrillar as in light chain amyloid or nonfibrillar as in light chain deposition disease. The reasons for these structural differences are still unknown. We characterized the myocardial deposits by immunohistochemical examination of sections and extraction and biochemical analysis of the tissue deposits in a patient (MCM) who died of myeloma and systemic light chain deposition disease. Amino acid sequence analysis of the extracted nonfibrillar MCM kappa-light chain reveals that it belongs to the L12a germline subset of the kappa(I) protein and contains five distinctive amino acid substitutions (three in the ***framework*** region III and two in the ***complementarity*** - ***determining*** region III) that have not been reported previously in the same positions in other kappa(I) light chains. The theoretically determined isoelectric point (pI 8.21) of the MCM light chain is high compared with the low isoelectric point of other Bence Jones proteins from subjects without light chain deposition disease. The diffuse binding to basement membranes and the high isoelectric point of the MCM kappa-light chain suggest electrostatic interaction as a possible mechanism of tissue deposition. The spatial locations of the five distinctive residues and a sixth ***rare*** substitution of the MCM protein modeled on the backbone structure of REI, a kappa(I)-soluble Bence Jones light chain of known three-dimensional structure, may be responsible for protein destabilization, partial unfolding, and aggregation leading to tissue deposition.

L12 ANSWER 23 OF 34 MEDLINE DUPLICATE 19
ACCESSION NUMBER: 96134428 MEDLINE
DOCUMENT NUMBER: 96134428 PubMed ID: 8527402
TITLE: Establishment and structural analysis of human mAb to the E2 component of the 2-oxoglutarate dehydrogenase complex generated from a patient with primary biliary cirrhosis.
AUTHOR: Fukushima N; Nakamura M; Matsui M; Ikematsu H; Koike K; Ishibashi H; Hayashida K; Niho Y
CORPORATE SOURCE: First Department of Internal Medicine, Faculty of Medicine, Kyushu University, Fukuoka, Japan.
SOURCE: INTERNATIONAL IMMUNOLOGY, (1995 Jul) 7 (7) 1047-55.
Journal code: 8916182. ISSN: 0953-8178.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199601
ENTRY DATE: Entered STN: 19960220
Last Updated on STN: 19960220
Entered Medline: 19960129

AB We established one Epstein-Barr virus-transformed B cell hybrid clone producing human mAb of the IgG class to the 2-oxoglutarate dehydrogenase complex (OGDC) for the first time from the peripheral B lymphocytes of a patient with primary biliary cirrhosis (PBC). This mAb, designated mAbM37GO37, specifically bound to OGDC and its dissociation constant with OGDC was calculated to be 3.70 x 10(-10) mol/l. mAbM37GO37 stained murine stomach/kidney cryostat sections in a ***typical*** immunofluorescence pattern of antimitochondrial antibody (AMA). Western blotting analysis revealed that mAbM37GO37 reacted with an E2 component of OGDC but not with other components of OGDC nor pyruvate dehydrogenase complex (PDC). Furthermore, mAbM37GO37 completely inhibited the enzymatic activity of OGDC. In order to determine the structure and genetic origin of anti-OGDC autoantibody, we cloned and sequenced the Ig heavy and light chain variable regions of mAbM37GO37. This mAb used the VHIII family member,

V3-7, and the V kappa IV family member. The amino acid difference between the expressed V genes of this mAb and respective putative germline genes was concentrated within the ***complementarity*** ***determining*** regions (***CDR***) rather than the ***framework*** regions (FR). The R:S mutation ratio was high in the ***CDR*** and low in the FR. These features suggested that the immune response to OGDC is similar to that to exogenous antigen, and that the heavy and light chain variable regions of the anti-OGDC antibody undergo somatic hypermutation through antigen-driven clonal selection. This human mAb to OGDC, which was established for the first time from a patient with PBC and characterized at the molecular level, would be a valuable tool to study the B cell autoepitopes of OGDC, to clone as yet undetermined full length cDNA encoding OGDC and to dissect the autoimmune response to mitochondrial antigens in PBC.

L12 ANSWER 24 OF 34 MEDLINE DUPLICATE 20
ACCESSION NUMBER: 95232114 MEDLINE
DOCUMENT NUMBER: 95232114 PubMed ID: 7716162
TITLE: Efficient generation of a reshaped human mAb specific for the alpha toxin of Clostridium perfringens.
AUTHOR: Tempest P R; White P; Williamson E D; Titball R W; Kelly D C; Kemp G J; Gray P M; Forster S J; Carr F J; Harris W J
CORPORATE SOURCE: Scotgen Biopharmaceuticals Inc., Aberdeen, UK.
SOURCE: PROTEIN ENGINEERING, (1994 Dec) 7 (12) 1501-7.
Journal code: 8801484. ISSN: 0269-2139.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-L25352; GENBANK-L25353
ENTRY MONTH: 199505
ENTRY DATE: Entered STN: 19950524
Last Updated on STN: 19990129
Entered Medline: 19950516
AB We have used the technique of antibody reshaping to produce a humanized antibody specific for the alpha toxin of Clostridium perfringens. The starting antibody was from a mouse hybridoma from which variable (V) region nucleotide sequences were determined. The ***complementarity*** - ***determining*** regions (CDRs) from these V regions were then inserted into human heavy and light chain V region genes with human constant region gene fragments subsequently added. The insertion of CDRs alone into human frameworks did not produce a functional reshaped antibody and modifications to the V region ***framework*** were required. With minor ***framework*** modifications, the affinity of the original murine mAb was restored and even exceeded. Where affinity was increased, an altered binding profile to overlapping peptides was observed. Computer modelling of the reshaped heavy chain V regions suggested that amino acids ***adjacent*** to CDRs can either contribute to, or distort, ***CDR*** loop conformation and must be adjusted to achieve high binding affinity.

L12 ANSWER 25 OF 34 MEDLINE DUPLICATE 21
ACCESSION NUMBER: 92113283 MEDLINE
DOCUMENT NUMBER: 92113283 PubMed ID: 1730882
TITLE: Franklin's disease: Ig gamma 2 H chain mutant BUR.
AUTHOR: Prelli F; Frangione B
CORPORATE SOURCE: Department of Pathology, New York University Medical Center, NY 10016.
CONTRACT NUMBER: AR 02594 (NIAMS)
SOURCE: JOURNAL OF IMMUNOLOGY, (1992 Feb 1) 148 (3) 949-52.
Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199202
ENTRY DATE: Entered STN: 19920308
Last Updated on STN: 19920308

Entered Medline: 19920219

AB The complete sequence of a gamma 2-H chain disease protein BUR is presented. This mutant, a dimer of a 348-residue chain, linked by four disulfide bridges, is composed of a complete V region, hinge, CH2, and CH3 domains. There is one deletion, the CH1 domain, which includes the cysteine residue bridging the H to L chain. Although the V region is encoded by the VHI and JHIII genes, it has several distinctions: methionine at position 11, two unique cysteine residues in the second ***complementarity*** ***determining*** region (CDR2), and three glycosylation sites, two of which are located in the CDR2 and CDR3 regions. These distinctive characteristics of BUR VH within the ***framework*** of a normal VHI may be affected by extensive somatic mutation or by a ***rare*** and previously unanalyzed VH gene.

L12 ANSWER 26 OF 34 MEDLINE DUPLICATE 22
ACCESSION NUMBER: 91287731 MEDLINE
DOCUMENT NUMBER: 91287731 PubMed ID: 1905783
TITLE: Immunoglobulin VH genes of the goldfish, Carassius auratus: a re-examination.
AUTHOR: Wilson M R; Middleton D; Warr G W
CORPORATE SOURCE: Department of Biochemistry and Molecular Biology, Medical University of South Carolina, Charleston 29425.
SOURCE: MOLECULAR IMMUNOLOGY, (1991 Apr-May) 28 (4-5) 449-57. Journal code: 7905289. ISSN: 0161-5890.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-D12728; GENBANK-D12729; GENBANK-D12730; GENBANK-D12731; GENBANK-D12732; GENBANK-D12733; GENBANK-X61312; GENBANK-X61313; GENBANK-X61314; GENBANK-X65266
ENTRY MONTH: 199108
ENTRY DATE: Entered STN: 19910825
Last Updated on STN: 19910825
Entered Medline: 19910807

AB Five VH-related genomic sequences from the goldfish, Carassius auratus, have been characterized. One of these sequences appeared to be a functional gene, and four to be pseudogenes. The main conclusions drawn from this study were that: (1) With minor exceptions, goldfish VH genes conform to the ***typical*** pattern of vertebrate VH genes in terms of their structure (they encode an intron-split hydrophobic leader, 3 ***framework*** and 2 ***complementarity*** - ***determining*** regions, and possess a ***typical*** 3' recombination signal sequence for VH to D joining) and regulatory sequences (possession of a ***typical*** upstream octameric promoter). (2) The sequences indicate that goldfish possess multiple families of VH sequences (at least three). Two of these families contain approximately 6 and 10 members, as judged from Southern blot hybridization experiments. (3) Goldfish VH gene families are distributed throughout the members of the species in the manner ***typical*** of that of VH families in other vertebrate species. Thus, this observation corrects the previous conclusion (Wilson et al., Proc. natn. Acad. Sci. U.S.A. 85, 1566-1570, 1988) that VH genes are discontinuously distributed in the goldfish population.

L12 ANSWER 27 OF 34 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1991:183735 CAPLUS
DOCUMENT NUMBER: 114:183735
TITLE: Chimeric immunoglobulins specific for p55 Tac protein of the interleukin-2 (IL-2) receptor
INVENTOR(S): Queen, Cary L.; Selick, Harold Edwin
PATENT ASSIGNEE(S): Protein Design Labs, Inc., USA
SOURCE: PCT Int. Appl., 48 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 4

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9007861	A1	19900726	WO 1989-US5857	19891228
W: AT, AU, BB, BG, BR, CH, DE, DK, FI, GB, HU, JP, KP, KR, LK, LU, MC, MG, MW, NL, NO, RO, SD, SE, SU				
RW: AT, BE, BF, BJ, CF, CG, CH, CM, DE, ES, FR, GA, GB, IT, LU, ML, MR, NL, SE, SN, TD, TG				
CA 2006865	AA	19900628	CA 1989-2006865	19891228
CA 2006865	C	20020820		
CN 1043875	A	19900718	CN 1989-109618	19891228
CN 1057013	B	20001004		
AU 9051532	A1	19900813	AU 1990-51532	19891228
AU 647383	B2	19940324		
ZA 8909956	A	19901031	ZA 1989-9956	19891228
EP 451216	A1	19911016	EP 1990-903576	19891228
EP 451216	B1	19960124		
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE				
JP 04502408	T2	19920507	JP 1990-503677	19891228
EP 682040	A1	19951115	EP 1995-105609	19891228
EP 682040	B1	19990825		
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE				
AT 133452	E	19960215	AT 1990-903576	19891228
ES 2081974	T3	19960316	ES 1990-903576	19891228
JP 11004694	A2	19990112	JP 1998-4334	19891228
RU 2126046	C1	19990210	RU 1989-4895847	19891228
EP 939127	A2	19990901	EP 1998-204240	19891228
R: AT, BE, CH, DE, ES, FR, GB, IT, LI, LU, NL, SE				
AT 183753	E	19990915	AT 1995-105609	19891228
ES 2136760	T3	19991201	ES 1995-105609	19891228
CA 2328851	C	20020813	CA 1989-2328851	19891228
DD 296964	A5	19911219	DD 1990-337159	19900117
DK 9101191	A	19910619	DK 1991-1191	19910619
NO 9102385	A	19910619	NO 1991-2385	19910619
SG 78258	A1	20010220	SG 1996-7855	19960413
DK 9800941	A	19980716	DK 1998-941	19980716
DK 174317	B1	20021202		
HK 1014718	A1	20000714	HK 1998-115967	19981228

PRIORITY APPLN. INFO.: US 1988-290975 A 19881228
US 1989-310252 A 19890213
CA 1989-2006865 A3 19891228
EP 1990-903576 A3 19891228
EP 1995-105609 A3 19891228
JP 1990-503677 A3 19891228
WO 1989-US5857 A 19891228

AB Methods for designing humanized Igs having .gtoreq.1 complementary detg. regions (CDRs) from a donor Ig and a ***framework*** region from a human Ig comprise 1st comparing the ***framework*** or variable region amino acid sequence of the donor Ig to corresponding sequences in a collection of human Ig chains and selecting as the human Ig .gtoreq. 1 homologous sequences from the collection. Each humanized Ig chain may comprise approx. .gtoreq.3 amino acids from the donor Ig in addn. to the CDRs, usually .gtoreq.1 of which is immediately ***adjacent*** to a ***CDR*** in the donor Ig. The heavy and light chains may each be designed by using any 1 or all 3 addnl. position criteria. When combined in an intact antibody, the humanized Igs of the invention will be substantially nonimmunogenic in humans and retain substantially the same affinity as the donor Ig to the antigen. The above method is applied to design and prodn. of the title chimeric antibodies. Thus, comparison of human antibody Eu and anti-Tac antibody sequences allowed design of genes for human-like light and heavy chains. Appropriate oligonucleotide segments were synthesized and annealed to form these genes, which were then used to construct plasmids pHuGTAC1 (for humanized heavy chain prodn.) and pHuLTAC (for humanized light chain prodn.). These 2 plasmids were transfected into mouse sp2/0 cells for prodn. of the title antibody. The secreted antibody bound to HUT-102 cells (which express the IL-2

receptor), but not to Jurkat T-cells (which do not express the IL-2 receptors). The humanized and original anti-Tac antibodies have approx. the same affinity (within 3-4-fold). In 51Cr-release antibody-dependent cell-mediated cytotoxicity assays using effector cell/target cell ratios of 30:1 or 100:1, anti-Tac antibody lysed <5% of target cells, while the humanized antibody lysed >20% of target cells.

L12 ANSWER 28 OF 34 MEDLINE DUPLICATE 23
ACCESSION NUMBER: 90375929 MEDLINE
DOCUMENT NUMBER: 90375929 PubMed ID: 2398280
TITLE: The BALB/c secondary response to the Sb site of influenza virus hemagglutinin. Nonrandom silent mutation and unequal numbers of VH and Vk mutations.
AUTHOR: Clarke S; Rickert R; Wloch M K; Staudt L; Gerhard W; Weigert M
CORPORATE SOURCE: Department of Microbiology and Immunology, University of North Carolina, Chapel Hill 27514.
CONTRACT NUMBER: AI-26844 (NIAID)
CA-06927 (NCI)
GM-20964 (NIGMS)
+
SOURCE: JOURNAL OF IMMUNOLOGY, (1990 Oct 1) 145 (7) 2286-96.
Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
OTHER SOURCE: GENBANK-M36740; GENBANK-M36741; GENBANK-M36742; GENBANK-M36743; GENBANK-M36744; GENBANK-M36745; GENBANK-M36746; GENBANK-M36747; GENBANK-M36748; GENBANK-M36749; GENBANK-M36750; GENBANK-M36751; GENBANK-M36752; GENBANK-M36753; GENBANK-M36754; GENBANK-M36755; GENBANK-M36756; GENBANK-M36757; GENBANK-M36758; GENBANK-M36759; GENBANK-M36760; GENBANK-M36761; GENBANK-M36762; GENBANK-M36763; GENBANK-M36764; GENBANK-M36765; GENBANK-M36766; GENBANK-M36767; GENBANK-M36880
ENTRY MONTH: 199010
ENTRY DATE: Entered STN: 19901122
Last Updated on STN: 19901122
Entered Medline: 19901017
AB We have determined the nucleotide sequences of the expressed VH and Vk genes from 13 secondary (2 degrees) hemagglutinin (HA) (Sb) specific hybridomas derived from a single mouse. These antibodies share an Id, H37-68 (68Id) that dominates the 2 degrees HA(Sb) response in this mouse, but is ***rare*** or absent from 2 degrees responses of other mice. We find that these antibodies derive from five clones. The H chains of these antibodies are encoded by a single VH gene joined to a variety of DH and JH genes. The length of ***complementarity*** - ***determining*** region (***CDR***) 3 and sequence of the D-J junction are restricted, suggesting selection on CDR3 of the H chain. The L chains are more diverse. In the presented examples, they are encoded by the Vk21C and Vk21E genes and a Vk9 gene, and are joined to Jk1, 2, or 4. Each antibody is extensively mutated. The nature and distribution of the mutations suggests that 68Id-producing cells have been selected by Ag, although there are differences regarding the domain (VH, Vk, or both) in which mutations were selected. The implications of these findings on the idiosyncratic nature of the 68Id antibody response to HA(Sb) are discussed. There are two unusual characteristics regarding somatic mutation in these hybridomas. Whereas the expressed VH and Vk21 genes appear to have accumulated mutations at a high rate (1 to 1.5 x 10(-3)/base pairs/division, the expressed Vk9 genes appear to have accumulated mutations at a 5 to 15-fold lower rate than the expressed VH genes in the same cells. There is also a surprisingly high number of parallel silent somatic mutations in the VH genes, of which all but one are clustered to a 28-bp region in ***framework*** region 2 and ***CDR*** 2-encoding segments. The probability that this could have

occurred by a random mutational process is essentially zero.

L12 ANSWER 29 OF 34 MEDLINE DUPLICATE 24
ACCESSION NUMBER: 90324610 MEDLINE
DOCUMENT NUMBER: 90324610 PubMed ID: 2115548
TITLE: Idiotope structure and genetic diversity in anti-streptococcal group A carbohydrate antibodies.
AUTHOR: Phillips N J; Davie J M
CORPORATE SOURCE: Department of Pathology, Washington University School of Medicine, St. Louis, MO 63110.
CONTRACT NUMBER: AI 15926 (NIAID)
AI07163-09 (NIAID)
SOURCE: JOURNAL OF IMMUNOLOGY, (1990 Aug 1) 145 (3) 915-24.
Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 199008
ENTRY DATE: Entered STN: 19901012
Last Updated on STN: 19901012
Entered Medline: 19900828

AB Three cross-reactive idiotopes(Id), termed IdX, Idl-1, and Id5, that are present on free L chains from murine anti-group A streptococcal carbohydrate antibodies have been mapped; these Id distinguish between products of three homologous V kappa genes. For each determinant, sequence analysis of anti-streptococcal group A carbohydrate antibody V domains yielded small numbers of amino acids invariably associated with Id expression. Flow micro-fluorimetry was used to isolate three Idl-1- spontaneous mutants of the Idl-1+ hybridoma GAC 39; all had single amino acid changes in the L chain at position 60 and 77, all retained other Id, and all bound group A carbohydrate. Computer modeling was used to examine spatial relationships between Id. A number of the conserved Id5 and IdX residues cluster in the L chain ***framework*** region 1 around the first back loop connecting strands of the beta pleated sheets, and overlap at residue 15 (Id5, proline; IdX, leucine). This overlap accords with the mutually exclusive expression of Id5 and IdX. The Idl-1 loss variants have mutations of residues 60 or 77 on ***adjacent*** back loops, approximately 7.5 and 14 A from residue 15. Competitive inhibition of anti-IdX and anti-Idl-1 binding to antibodies expressing both Id can be attributed to steric hindrance. The ***framework*** back loops may be favored sites for cross-reactive Id expressed by products of a single V region gene. Idl-3a, an individual Id not associated with use of a particular gene segment, has been localized in part to residue 31 (***hypervariable*** region 1) of the H chain.

L12 ANSWER 30 OF 34 MEDLINE DUPLICATE 25
ACCESSION NUMBER: 89035545 MEDLINE
DOCUMENT NUMBER: 89035545 PubMed ID: 3141511
TITLE: Genetics of the phosphocholine-specific antibody response to Streptococcus pneumoniae. Germ-line but not mutated T15 antibodies are dominantly selected.
AUTHOR: Claflin J L; Berry J
CORPORATE SOURCE: Department of Microbiology and Immunology, University of Michigan Medical School, Ann Arbor 48109.
CONTRACT NUMBER: AI12533 (NIAID)
AI23755 (NIAID)
SOURCE: JOURNAL OF IMMUNOLOGY, (1988 Dec 1) 141 (11) 4012-9.
Journal code: 2985117R. ISSN: 0022-1767.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 198812
ENTRY DATE: Entered STN: 19900308
Last Updated on STN: 19970203
Entered Medline: 19881220

AB The role that somatic mutations play in the phosphocholine-specific, antibody response to Streptococcus pneumoniae was examined by studying sets of hybridomas from different individual mice. As expected most of the cell lines were from the T15 anti-phosphocholine family and were not encoded by the v1 gene of the T15 VH family and V kappa 22. A minority of antibodies were from the M603 (v1/V kappa 8) and M511 (v1/V kappa 24) families. Three additional antibodies were encoded by the v11 gene of the T15 family; two were paired with a V lambda and the other with a V kappa 1 gene. In vitro binding studies showed that T15- and M603-like antibodies had the highest affinity for S. pneumoniae. Complete sequencing of the VH and VL mRNA from 25 of the hybridomas revealed somatic mutations in 11 of the antibodies. A total of 17 independently derived T15 positive cell lines were studied in detail, six of these were mutated. These mutations were scattered throughout the V regions and the replacement to silent ratio was ***typical*** of that for ***framework*** regions. Statistical evaluation of the placement of mutations showed that there was a slight but significantly decreased frequency of mutations in ***complementarity*** ***determining*** regions. Comparisons of mutated and unmutated T15-related antibodies showed that mutations caused a decrease in binding to S. pneumoniae in every case. These results argue that the optimal specificity for this molecular form of phosphocholine is encoded in the germline and that Ag-driven events favor selection of B cells expressing these germ-line encoded antibodies.

L12 ANSWER 31 OF 34 MEDLINE
ACCESSION NUMBER: 85140192 MEDLINE
DOCUMENT NUMBER: 85140192 PubMed ID: 2983316
TITLE: Complete nucleotide sequences of three VH genes in Caiman, a phylogenetically ancient reptile: evolutionary diversification in coding segments and variation in the structure and organization of recombination elements.
AUTHOR: Litman G W; Murphy K; Berger L; Litman R; Hinds K; Erickson B W
CONTRACT NUMBER: CA 08748 (NCI)
GM 32106 (NIGMS)
GM 32622 (NIGMS)
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1985 Feb) 82 (3) 844-8.
Journal code: 7505876. ISSN: 0027-8424.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: GENBANK-M12768; GENBANK-M12769; GENBANK-M12770
ENTRY MONTH: 198503
ENTRY DATE: Entered STN: 19900320
Last Updated on STN: 19970203
Entered Medline: 19850327

AB Complete nucleotide sequences are described for three caiman (Caiman crocodylus crocodylus) immunoglobulin VH genes (C3, E1, and G4) that hybridize with a murine VH probe. The E1 and G4 genes are physically linked (intergenic distance, approximately equal to 6.5 kilobases) in the same transcriptional orientation but are not directly contiguous with the C3 gene. When the coding segments, including both ***framework*** and ***complementarity*** - ***determining*** regions, of these genes and the murine probe sequences are compared by metric analysis, it is apparent that the caiman genes are only slightly more related to each other than to the mammalian sequence, consistent with significant preservation of nucleotide sequence over an extended period of phylogenetic time. Based on the presence of transcriptionally critical 5' sequences and the absence of terminator codons, frameshift mutations, or other recognizable alterations, the genes do not appear to be pseudogenes. The E1 gene, however, is distinguished from other VH genes because (i) the spacer region within the 3' recombination signal sequence is 12 base pairs, ***typical*** of VK genes but not of VH genes, which possess 22- to 23-base-pair spacers and (ii) a near-perfect VH recombination signal sequence is present within the intervening sequence that splits the

segment encoding the leader. These studies establish VH gene multiplicity in a species that arose prior to mammalian radiation and provide a description of differences in the configuration and location of recombination elements associated with an otherwise potentially functional gene.

L12 ANSWER 32 OF 34 MEDLINE DUPLICATE 26
ACCESSION NUMBER: 82105968 MEDLINE
DOCUMENT NUMBER: 82105968 PubMed ID: 6798566
TITLE: Mouse immunoglobulin coding sequences for the heavy-chain variable region arose as repeats of the two short building blocks.
AUTHOR: Ohno S; Kato K; Hozumi T; Matsunaga T
CONTRACT NUMBER: RO1 ATT 5620 (NCCAM)
SOURCE: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1982 Jan) 79 (1) 132-6.
Journal code: 7505876. ISSN: 0027-8424.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198203
ENTRY DATE: Entered STN: 19900317
Last Updated on STN: 19900317
Entered Medline: 19820322
AB The coding sequence for the 97-amino-acid-residue-long immunoglobulin heavy-chain variable (VH) regions of the mouse apparently arose as repeats of the two short building blocks. Three of the recognizable copies of the one 21-base-long prototype sequence A-C-T-G-G-A-T-A-T-G-A-C-C-T-G-G-A-G-T-G-G are invariably found to occupy the fixed positions within the 5' half of each VH coding sequence. Interestingly, the first and third copies specify the relatively invariant regions represented by the 7th to 13th and 41st to 47th amino acid residues (the first and second ***framework*** regions), whereas the second copy specifies the first ***hypervariable*** region (31st to 35th amino acid residues). These copies maintain at least 57.2% (12 out of 21) base sequence homology to the above-noted prototype building block. Base sequences of the other 14- to 15-base-long prototype building block differ from each other by as much as 60% between individual VHS. Yet one of its copies invariably occupies the terminal region of each VH coding sequence, thus specifying the very invariant third ***framework*** region. Other copies occupy unfixed positions in the VH and its attendant hydrophobic leader coding sequence as well as in ***adjacent*** noncoding sequences. The homology thus revealed between the VH coding sequence and its ***adjacent*** noncoding sequences suggests their concordant evolution.

L12 ANSWER 33 OF 34 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.DUPLICATE 27
ACCESSION NUMBER: 1979:232284 BIOSIS
DOCUMENT NUMBER: BA68:34788
TITLE: EQUILIBRIUM AND KINETIC ASPECTS OF THE INTERACTION OF ISOLATED VARIABLE AND CONSTANT DOMAINS OF LIGHT CHAIN WITH FC-PRIME FRAGMENT OF IMMUNO GLOBULIN G.
AUTHOR(S): KLEIN M; KORTAN C; KELLS D I C; DORRINGTON K J
CORPORATE SOURCE: DEP. IMMUNOL., TORONTO WEST. HOSP., TORONTO, ONT., CAN.
SOURCE: BIOCHEMISTRY, (1979) 18 (8), 1473-1481.
CODEN: BICHAW. ISSN: 0006-2960.
FILE SEGMENT: BA; OLD
LANGUAGE: English
AB The noncovalent interaction of [human] .kappa. L chains and fragments corresponding to their variable (V.kappa.) and constant (C.kappa.) domains with Fd' fragments of immunoglobulin [Ig] G was studied by UV difference spectroscopy in 4 mM sodium acetate buffer (pH 5.4). At the concentrations used, the intact .kappa. chains, the V.kappa. fragments and Fd' existed as monomer-dimer equilibrium mixtures, and the C.kappa. fragments were monomeric. Characteristic red-shifted difference spectra were obtained when V.kappa. and C.kappa. were recombined separately with Fd', suggesting

that aromatic chromophores were transferred to a nonpolar environment. When these spectra were summed, significant differences were noted when compared to the spectrum obtained when the parent .kappa. chain was recombined with Fd'. When V.kappa. and C.kappa. were simultaneously bound to Fd', the spectrum was identical with that observed with the intact .kappa. chain. When V.kappa. or C.kappa. fragments were recombined with a binary complex formed with Fd' and the complementary domain fragment, significantly different spectra were observed. Equilibrium binding curves were constructed from the spectral data indicating a 1:1 binding between Fd' and V.kappa. and C.kappa.. The fragments bound to Fd' with substantially lower affinity than that for the parent .kappa. chain. The affinity of V.kappa. was enhanced in the presence of C.kappa.. The time dependence of the spectral changes was used to determine the rates of domain association. All reactions were 2nd order. The forward rate constant for the binding of V.kappa. to Fd' was the same as for the intact .kappa. chain and was not affected by the presence of C.kappa. (.apprx. 200 M-1 s-1). C.kappa. was bound much more slowly (.apprx. 10 M-1 s-1) although the rate was significantly increased in the presence of V.kappa. (.apprx. 70 M-1 s-1). Although the C.kappa. fragments from several different .kappa. chains bound to Fd' in a similar way, only V.kappa. from the autologous .kappa. chain showed significant affinity for Fd'. In the presence of C.kappa. this specific interaction was no longer apparent and heterologous. V.kappa. fragments were capable of binding to Fd'. The high-affinity association of Fd' and .kappa. chain apparently derives from a combination of relatively weak interactions involving pairs of domains (i.e., VH-V.kappa. and C.gamma.1-C.kappa.), and the binding of l .kappa.-chain domain to Fd' induces a conformational change in the ***adjacent*** domain in Fd', thus modifying its reactivity toward the complementary .kappa.-chain domain. The specific association of autologous V.kappa. and VH may be due to quaternary interactions betwen ***hypervariable*** sequence regions or substitutions in the ***framework*** of the variable regions.

L12 ANSWER 34 OF 34 MEDLINE DUPLICATE 28
ACCESSION NUMBER: 77038198 MEDLINE
DOCUMENT NUMBER: 77038198 PubMed ID: 824717
TITLE: Complete amino acid sequence of the variable domains of two human IgM anti-gamma globulins (Lay/Pom) with shared idiotypic specificities.
AUTHOR: Capra J D; Klapper D G
SOURCE: SCANDINAVIAN JOURNAL OF IMMUNOLOGY, (1976) 5 (6-7) 677-84. Journal code: 0323767. ISSN: 0300-9475.
PUB. COUNTRY: Norway
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 197701
ENTRY DATE: Entered STN: 19900313
Last Updated on STN: 19900313
Entered Medline: 19770103

AB On the basis of extensive shared idiotypic specificities, two human IgM anti-gamma-globulins (Lay/Pom) were selected for complete amino acid sequence analysis of their variable domains. Previous studies on the variable regions of the heavy chains of these proteins had shown but eight amino acid differences, only one of which was within a ***complementarity*** - ***determining*** ***hypervariable*** region. The complete amino acid sequence of the variable regions of the light chains of these two proteins is the subject of this report. Protein Lay is a ***typical*** VchiI protein with only five ' ***framework*** ' differences when compared with protein Roy. Protein Pom is best classified as a Vchill, but in the ' ***framework*** ' there are 16 differences between it and protein Ti. Although there are extensive differences in the first ***hypervariable*** region, the second and third light-chain ***hypervariable*** regions have an identical sequence. The finding of two identical light-chain and two identical heavy-chain ***hypervariable*** regions in these two proteins, which were selected on the basis of their combining specificities and their

idiotypic cross-reactions, strongly implicates ***hypervariable*** regions in the constitution of the idiotypic determinants and the antibody combining site. Additionally, the finding of identical ***hypervariable*** regions in light chains of different V-region subgroups fulfills a prediction of the gene-interaction concept of antibody variability.

=> d his

(FILE 'HOME' ENTERED AT 14:46:16 ON 14 JUL 2003)

FILE 'MEDLINE, EMBASE, BIOSIS, CAPLUS' ENTERED AT 14:46:28 ON 14 JUL 2003

L1 11791 S HUMANIZED OR RESHAPED OR HUMANIZ### OR RESHAP###
L2 33010 S CONSENSUS SEQUENCE
L3 16 S L1 AND L2
L4 14 DUP REM L3 (2 DUPLICATES REMOVED)
L5 183577 S FRAMEWORK
L6 592 S L1 AND L5
L7 1432710 S ADJACENT OR TYPICAL OR RARE
L8 18 S L6 AND L7
L9 7 DUP REM L8 (11 DUPLICATES REMOVED)
L10 20681 S CDR OR HYPERVARIABLE OR (COMPLEMENTARITY DETERMINING)
L11 94 S L5 AND L10 AND L7
L12 34 DUP REM L11 (60 DUPLICATES REMOVED)